SYNTHESIS AND CHARACTERISATION OF MALTOSIDES AND LACTOSIDES BASED ON ALCOHOL MIXTURES OBTAINED FROM PALM OIL AND PALM KERNEL OIL.

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ABSTRACT

The headgroup of these glycolipids, which consist of a dissacharide i.e. maltose and lactose, provide good hydrophilicity. These glycolipids are comparable to alkyl polyglucosides (APGs), a common non-ionic surfactant used for detergents, particularly in the cosmeceutical industry. APGs are limited to high lipophilicity due to their low degree of polymerisation (~1.5) and their chemical diverse compositions make them unsuitable for bio-related applications. Technical grade glycolipids based on alcohol mixtures from reduced palm oil and palm kernel oil were investigated to explore their application as surfactants. The technical grade glycolipids were synthesized using a method that produces "clean" anomeric mixtures with no significant non-glycoside contamination, making them suitable for biotechnology application. The investigations involved thermotropic and lyotropic behaviour of the glycolipids mixtures. Two major effects were investigated separately; stereochemical effect and chain effect. Studies on stereochemical effects were performed on dodecyl (C_{12}) model compounds, comprising various α/β ratios based on different synthetic approaches. The chain effect study was devided in two categories; chain length and chain inhomogeneity. The clearing transition temperatures of palm oil and palm kernel oil maltosides are influenced by the unsaturated compounds while the lactosides' are more complicated. High unsaturation in palm oil maltosides showed a cubic phase but the palm oil and palm kernel oil lactosides have no significant difference in their lyotropic phase behaviour.

ABSTRAK

Glikolipid gred teknikal terdiri daripada campuran alkohol hasil daripada minyak sawit dan minyak kernel sawit dikaji untuk mengetahui aplikasinya sebagai surfaktan. Kumpulan kepala glikolipid ini terdiri daripada disakarida iaitu maltos dan lactos, memberikan sifat hidrofilik yang baik. Glikolipid ini menandingi alkil poliglukosida (APGs), surfaktan bukan ionik yang biasa digunakan untuk detergen khasnya dalam industri kosmesiutikal. APGs terhad kepada lipofilisiti yang tinggi disebabkan oleh darjah pempolimeran yang rendah (~1.5) dan komposisi kimia yang diversi membuatkan ia kurang sesuai untuk aplikasi biologi. Glaikolipid gred teknikal ini disintesis menggunakan cara yang menghasilkan campuran anomerik yang "bersih" tanpa pencemaran bahan bukan glaikosida, menjadikan ia sesuai untuk aplikasi bioteknologi. Kajian melibatkan sifat termotropik dan liotropik campuran glaikolipid ini. Dua pengaruh besar dikaji berasingan iaitu pengaruh stereokimia dan pengaruh rantai. Pengaruh stereokimia dikaji menggunakan model bahan dodesil (C_{12}) yang mengandungi pelbagai nisbah α/β hasil daripada perbezaan cara sintesis. Pengaruh rantai pula dibahagi kepada dua kategori; panjang rantai dan inhomogeniti rantai. 'Clearing transition temperature' maltosida sawit dan kernel sawit dipengaruhi oleh sebatian taktepu sementara sifat laktosida lebih kompleks. Ketaktepuan yang tinggi dalam maltosida sawit menunjukkan fasa kubik manakala laktosida sawit dan kernel sawit tidak menunjukkan perbezaan ketara dalam sifat fasa liotropik.

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Ac_2O	Acetic anhydride
AG	Alkyl glucoside
APG	Alkyl polyglucoside
Cello	Cellobioside
CMC	Critical micelle concentration
DP	Degree of polymerisation
DSC	Differential scanning calorimetry
et al.	*et alii*; and others
g	Glass phase
H_I	Normal hexagonal
H_{II}	Reverse hexagonal
HLB	Hydrophile-lipophile balance
I	Discontinuous cubic
i.e.	*id est*; that is
IV	Iodine value
Lacto	Lactoside
L_I	Micellar solution
L_{II}	Reverse micellar solution
L_α	Lamellar
Malto	Maltoside
MeOH	Methanol

NaOAc	Sodium acetate
NaOMe	Sodium methoxide
NMR	Nuclear magnetic resonance
OPM	Optical polarising microscopy
PEG	Polyethylene glycol
PKO	Palm kernel oil
PO	Palm oil
Q	Cubic
ROH	Alcohol
TLC	Thin layer chromatography
V	Bicontinuous cubic

1.1 Surfactants

Surfactant is a short form for **surf**ace **act**ive **a**ge**nt**. It is a wetting agent which lowers interfacial tensions. Surfactants have been used as active ingredients in detergents for many years. Applications of surfactants vary from emulsifiers to demulsifiers, detergent foaming agents, wetting agents, softeners and many more.

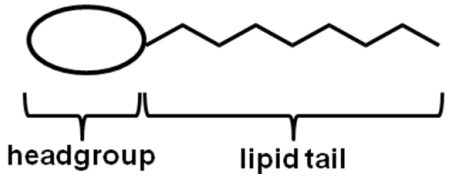


Figure 1.1 Surfactant structure

A surfactant consists of two regions, i.e. a hydrophilic headgroup and a hydrophobic lipid tail, which together act as amphiphile. When in contact with solvent (organic or inorganic, but usually water) the presence of two opposing tendencies will lead to self-assembled aggregates, whose precise structures are influenced by the concentrations and temperatures.

1.2 Surfactant Self-Assembly

Surfactants are characterised by their physicochemical properties. In order to have an optimum performance for a specific application, a surfactant is selected based on these properties, including phase behaviour and interfacial properties. Studies of these properties are relevant to understand the surfactant application as emulsifier.

For example, a phase diagram shows different liquid crystal phases, or self-assemblied structures, occurring at specific combinations of temperature and composition. All surfactants form micelles. Besides must also exhibit liquid crystal phases. There are

various lyotropic liquid crystalline phases exhibited by surfactants, such as the lamellar, hexagonal (or columnar) and cubic (bicontinuous or discontinuous) phases. They are discussed in the subtopics below.

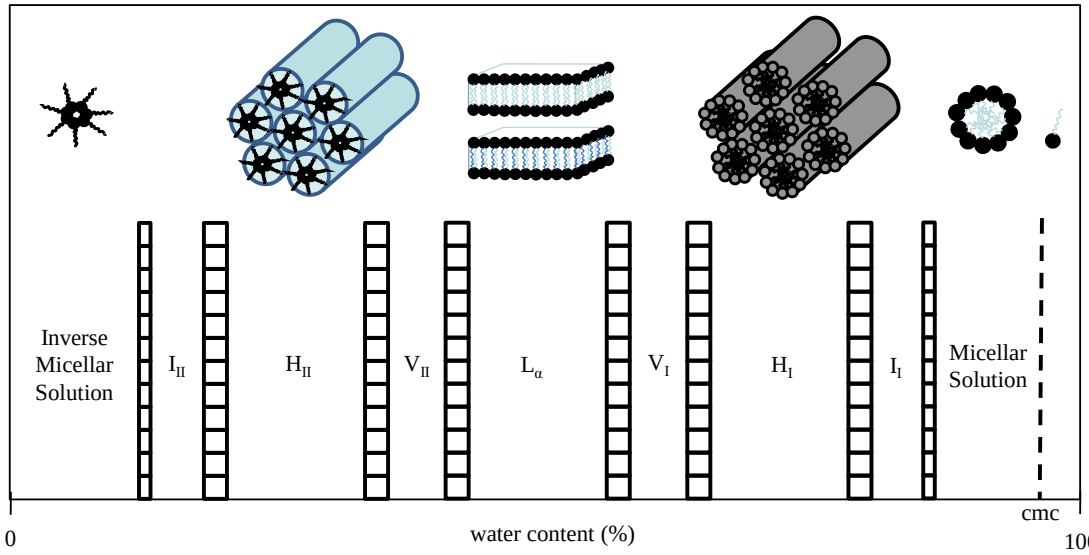


Figure 1.2 Principle phase behaviour of amphiphilic compounds (Seddon et al., 2001)

Surfactant molecules tend to accumulate at interfaces to lower the free energy and results in adsorption. Therefore they lower the surface tension of both media, thus enabling the formation of emulsions and suspensions. Common interfaces are water-air and water-oil.

Surface and interfacial activity of surfactant solutes are reflected by the critical micelle concentration (CMC). While there are many ways to determine the CMC, surface tension measurement is the simplest method and most commonly used. Factors that can affect this activity are the surfactant structure, co-surfactant and possible effect of electrolytes.

1.2.1 Lamellar phase

In the lamellar phase (figure 1.3), also known as neat soap phase, molecules arrange in multiple bilayers with water in between the bilayers. This molecular arrangement is similar to the thermotropic smectic A phase. The lamellar layer spacing is larger than the smectic layer because of the hydrogen bonded water molecules around the headgroup (Sakya and Seddon, 1997).

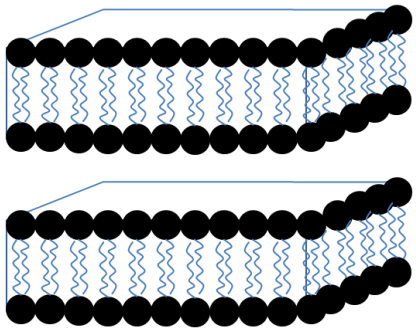


Figure 1.3 Lamellar phase

1.2.2 Hexagonal phase

The hexagonal phase (figure 1.4), or middle soap phase, occurs when micelles form hexagonal structure due to high concentration. Two types of hexagonal phase can be observed; the normal hexagonal (H_I) and the reverse hexagonal (H_{II}) phase. H_I is characterized by a continuous hydrophilic domain, and usually forms at high water content, whereas H_{II} is observed in non polar organic solvents. In this case the continuous phase is hydrophobic. Both phases give the same texture under OPM. They are rather viscous compared to the lamellar phase (Syed Husan, Rowe and Tiddy, 2001).

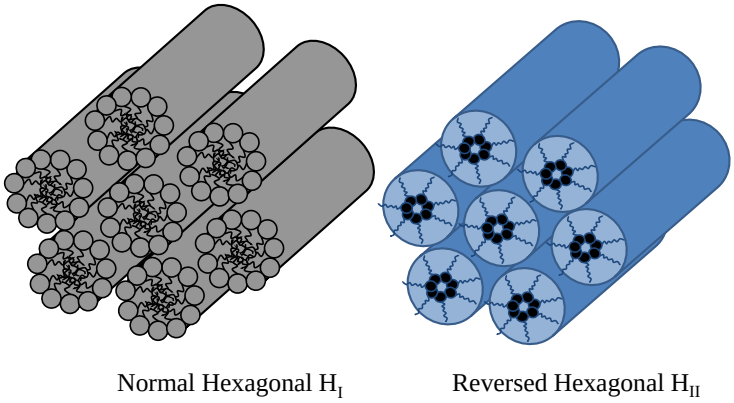


Figure 1.4 Hexagonal packing

1.2.3 Bicontinuous cubic phase

The bicontinuous cubic phase (V) is known as 'viscous isotropic phase' since it does not show any texture under OPM (optical polarizing microscope). This is due to the complex structure involving a continuous 3-dimensional networks. Nevertheless this phase is easily distinguished from a micellar solution because of its high viscosity. Usually a cubic phase is detected between a lamellar and a hexagonal phase. Apart from the bicontinuous cubic phase, there is also a discontinouos cubic phase (I), where small spherical micelles form primitive cubic or body and face centered lattices. Both, the bicontinuous and the discontinuous cubic phases are classified based on symmetry, two classes for each normal and reverse phase. Among the I phase structures observed are Pm3n, Im3m and Fm3m while V phases' are Pn3m, Im3m and Ia3d (figure 1.5).

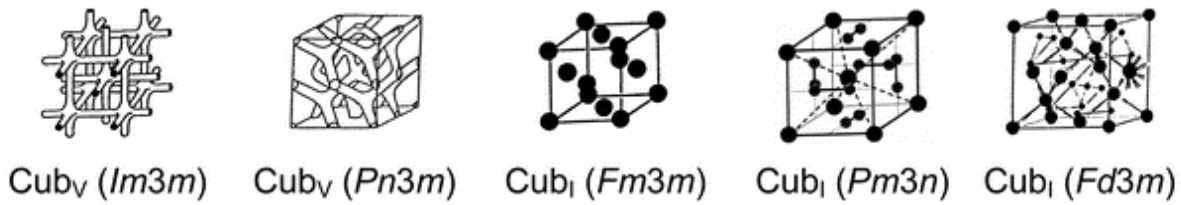


Figure 1.5 Cubic packing (Tschierske, 2001)

1.3 Surfactant Resources

A surfactant consists two domains, i.e. a lipid (hydrophobic) and a hydrophilic. Each domain is derived from different resources. The lipid domain is mainly an alkyl chain or a polymer whereas the hydrophilic domain varies from ionic to nonionic material, which categorise the surfactant type.

The lipid tail is normally based on natural fatty acids, petroleum fractions or relatively short synthetic polymers. However, due to environmental issues, petroleum source chemicals are more and more replaced by natural feedstock. These natural resources are biodegradable, (much) less flammable, harmless to the environment and, most importantly, renewable. One of the most common natural resource for fatty acids nowadays is palm oil.

1.3.1.1 Palm oil

Palm oil has been the most produced oil in the world for many years. In 2007, palm oil dominates the world oils and fats production with 38.2 million tons from 154 million tons which is 25% of the global productions (Oil World, March 2008). Malaysia is one of the largest producers in the world by producing about 40% of the world's palm oil in 2007 (Malaysian Palm Oil Board, 2007) (figure 1.6).

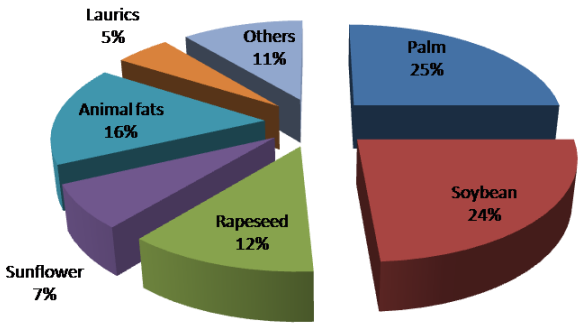


Figure 1.6 World oils and fats production 2007 (*Oil World, March 2008*)

The palm fruit is a unique crop since it produces two types of oils; palm oil from the mesocarp and palm kernel oil from the kernel (figure 1.7). These two oils have different fatty acid compositions. The palm oil contains of almost an equal amount of saturated and unsaturated fatty acids, which is similar to the chemical composition of tallow. Palm kernel oil, on the other hand, resembles coconut oil, containing predominantly saturated components. Detailed fatty acid compositions of these four oils are shown in table 1.1.

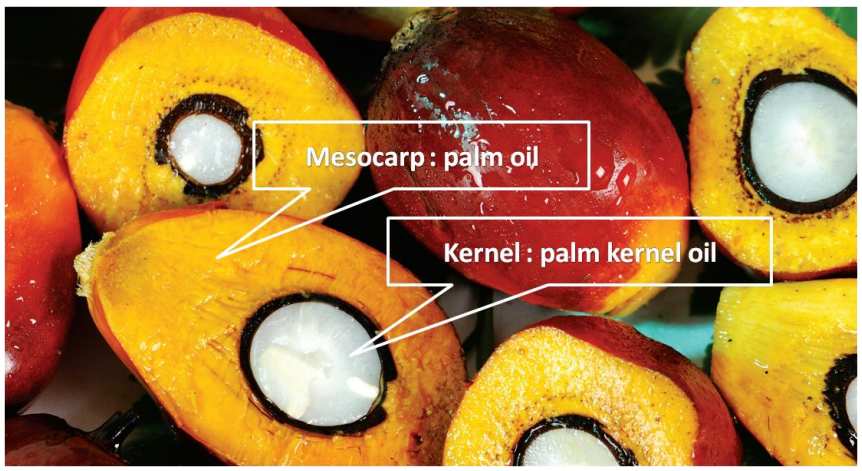


Figure 1.7 Palm oil fruit (image from MPOC, 2007)

Soap, detergents and personal care products are the most common industrial products of fatty acids. Nowadays most of the resource of fatty acids are produced from palm oil

and palm kernel oil. Before, tallow and coconut oils were the largest resources of fatty acids for C_{16}/C_{18} and C_{12}/C_{14} alkyl chains. However, due to similarities in the properties of the oils, as well as availability and price, palm oil and palm kernel oil have become the best fatty acid resources.

Table 1.1 Comparison of fatty acid composition of palm oil, tallow, palm kernel oil and coconut oil (de Vries, 1984)

	Fatty acids composition (%)					
	C12	C16	C18	C20	C18:1	C18:2
Palm Oil	0.1	43.7	4.4	0.3	39.9	10.3
Tallow		25.0	21.5	0.5	42.0	3.0
Palm kernel oil	48.0	7.7	1.7		15.6	2.7
Coconut oil	48.2	8.0	3.8		5.0	2.5

1.4 Surfactant Types

The hydrophilic headgroup can be charged or non-charged. Depending on the nature of the headgroup, surfactants are classified as anionic, cationic, zwitterionic and nonionic surfactants.

1.4.1 Anionic surfactants

An anionic surfactant has a negatively charged headgroup. It is used most frequently in the detergent industry (Schmitt, 1992). It has high foaming tendency and can remove soil more effectively than other surfactants. However, detergent builders are needed as

the surfactant is sensitive to hard water, because calcium and magnesium (in hard water) will partially deactivate the surfactant. Calcium and magnesium ions will form precipitates with fatty acids which are not water soluble thus leaving 'dirt' on the fiber. Anionic headgroups are mostly consisting of sulfates, carboxylates, phosphates and sulfonates (figure 1.8). Commercial anionic surfactants are sodium dodecyl sulfate (SDS), also known as sodium laureth sulfate (SLES), sodium perfluorooctanoate (PFOA or PFO) and sodium perfluorooctanesulfonate (PFOS) (figure 1.9).

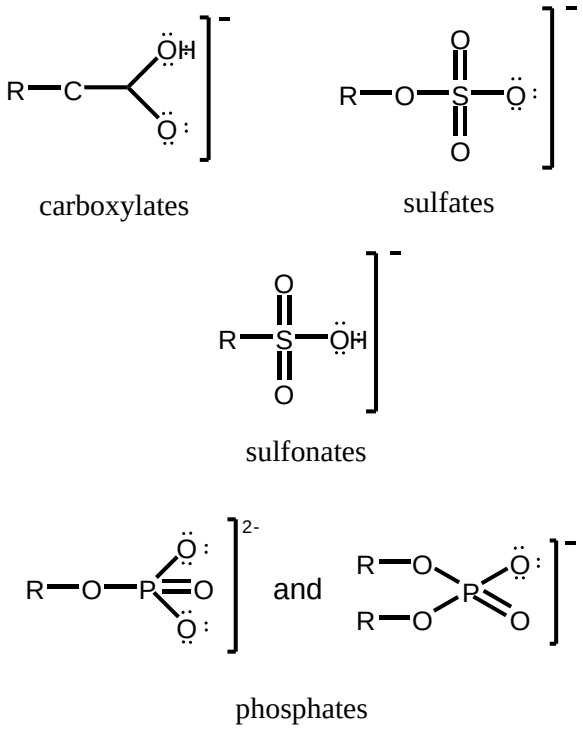
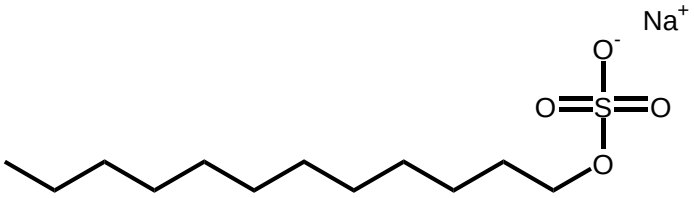
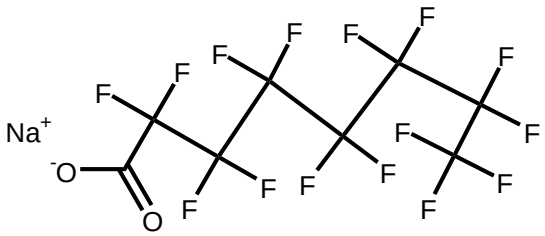


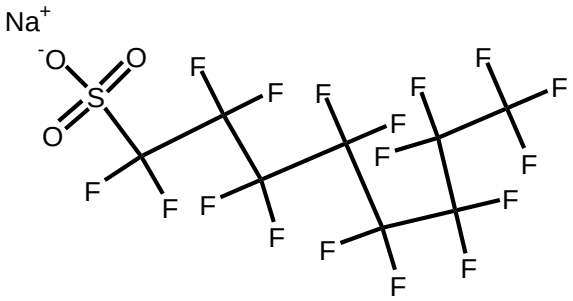
Figure 1.8 Anionic headgroups



Sodium dodecyl sulfate (SDS) or
Sodium laureth sulfate (SLES)



Sodium perfluorooctanoate
(PFOA or PFO)

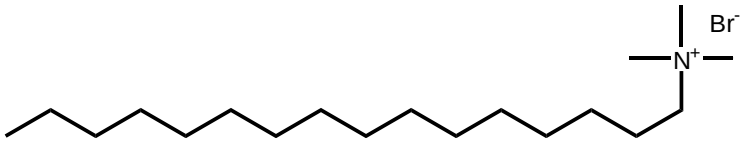


Sodium perfluorooctanesulfonate
(PFOS)

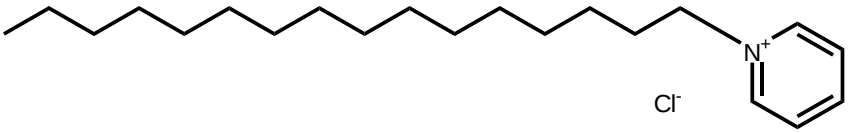
Figure 1.9 Commercial anionic surfactants

1.4.2 Cationic surfactants

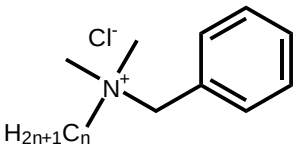
A cationic surfactant possesses a positively charged headgroup. It is mostly applied in the pharmaceutical and the pesticide industries. It is usually not suitable as detergent, because it requires low pH and is inefficient for cleaning at neutral pH. The headgroup is based on ammonium cations. Cetyl trimethylammonium bromide (CTAB), cetylpyridinium chloride and benzalkonium chloride (BAC) are few examples of commercial cationic surfactants (figure 1.10).



Cetyl trimethylammonium bromide



Cetylpyridinium chloride

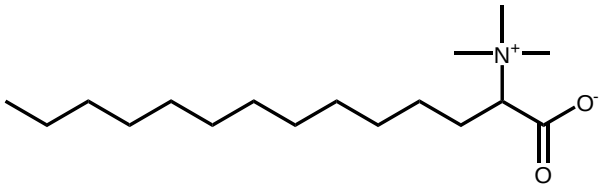


Benzalkonium chloride

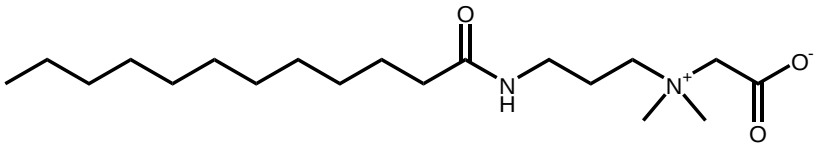
Figure 1.10 Commercial cationic surfactants

1.4.3 Zwitterionic surfactants

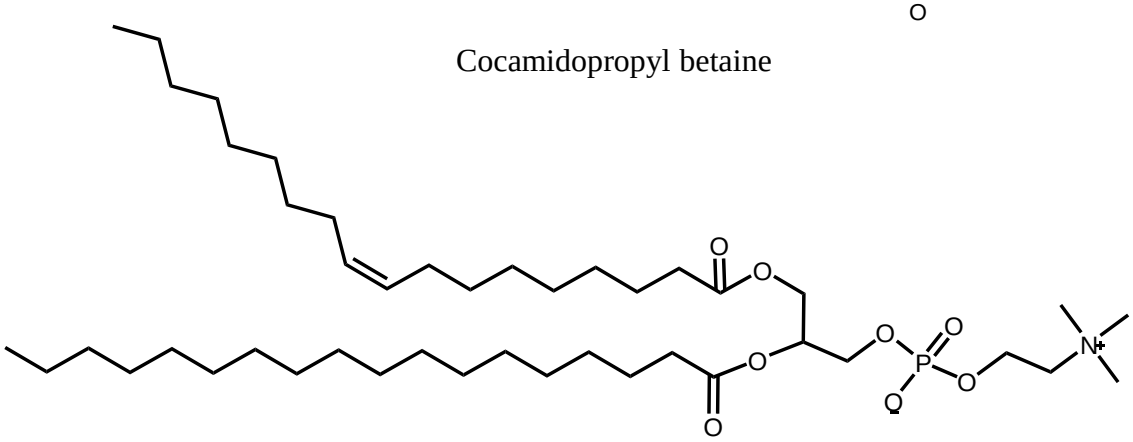
A zwitterion has two opposite charges; a positive (cationic) and a negative (anionic) but it is electrically neutral since the total net charge is zero. The zwitterionic surfactant can form anionic, cationic and non-ionic in aqueous solution depend on the pH. It is widely used in personal care products due to its excellent dermatological properties. Compared to the anionic surfactant, a zwitterionic is milder to the skin and low in eye irritation (Holmberg, 2003). Lecithin is a natural zwitterionic surfactant, while dodecyl betaine and cocamidopropyl betaine are synthesized from natural resources (figure 1.11).



Dodecyl betaine



Cocamidopropyl betaine



Lecithin

Figure 1.11 Zwitterionic surfactants

1.4.4 Nonionic surfactants

A nonionic surfactant has non-charged headgroup. Therefore, it does not dissociate to produce ions in water. This surfactant is used mainly for detergents and personal care products. It exhibits good cold water solubility, is a low-foamer and has a low CMC. Due to that it makes cleaning effective at low concentration (Schick, 1987). Unlike anionics, nonionics are more tolerant towards water hardness. Therefore no detergent builders are needed (Schmitt, 1992).

There are two major types of nonionic surfactants i.e. polyethylene glycol (PEG) based products and glycolipids. Figure 1.12 shows the classification of nonionic surfactants.

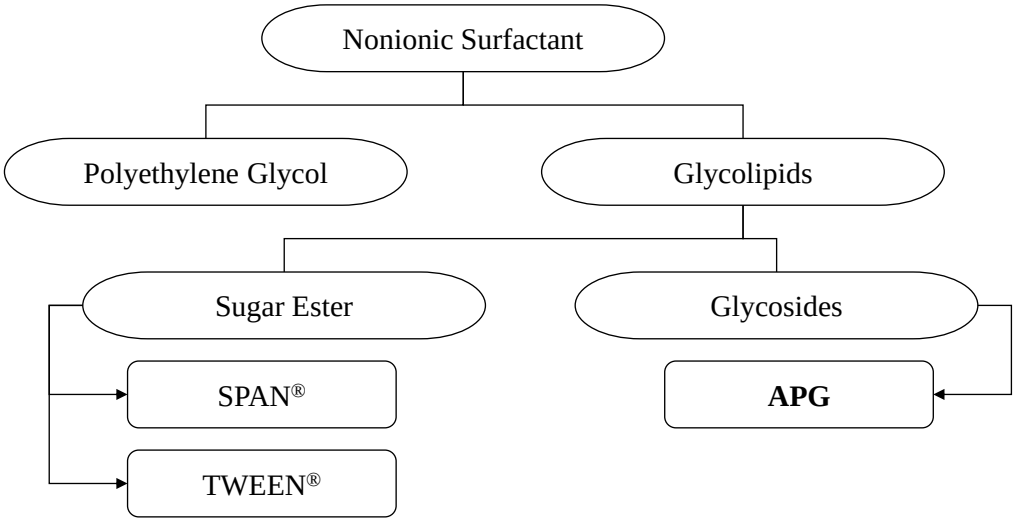


Figure 1.12 Nonionic surfactants classification

SPAN®, a commercial surfactant, consists of a monosaccharide sugar headgroup only and has an overbalanced hydrophobic region. Generally monosaccharides are applied due to purification difficulties and process selectivity. In order to increase the HLB (hydrophile-lipophile balance), ethylene glycol oligomers are added to the sugar hydroxyl groups, thus TWEEN® is produced. However these surfactants are easily saponified under mild basic condition, making them unsuitable to be used for liposomes, if alkali or acid treatment is required during formulation. Moreover they are also prone to microbial activity.

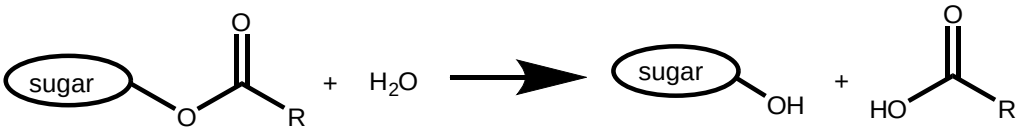
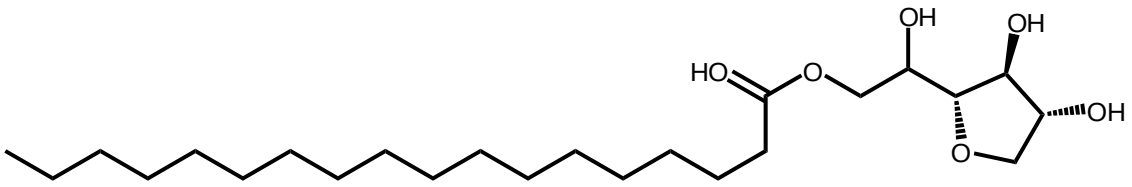


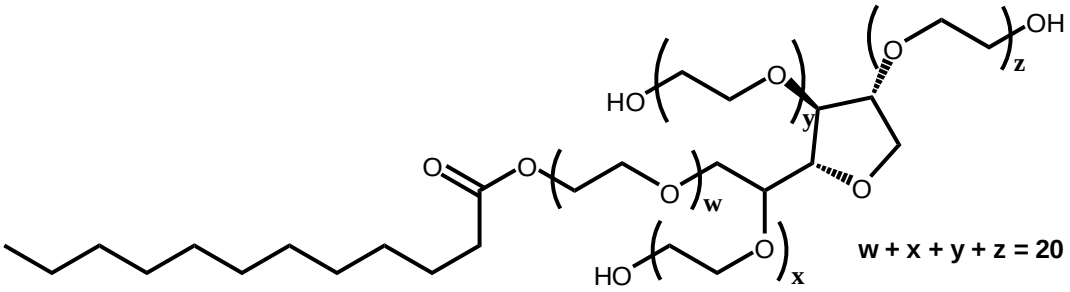
Figure 1.13 Saponification reaction of ester

APG, or alkyl polyglucoside, belongs to the glycoside group. It is widely used as emulsifier in the cosmeceutical industry due to high skin compatibility. It is chemically more stable than sugar esters, but has high a HLB due to the long alkyl chain that cannot be balanced by the sugar headgroup. Therefore, it has similar limitation as

SPAN® because the average size of the sugar is low (~1.5) (Balzer and Lüders, 2000). Besides, APG consists of a complex mixture of different oligomers as well as regio- and stereoisomers which makes it not suitable for drug delivery applications. This is because every chemical to be use in drug has to undergo toxicology and biological activity tests. Therefore a mixture is unfavourable, since all components have to be analysed separately and separation and testing of each compound is very expensive.



SPAN 60



TWEEN 20 (one possible structure)

Figure 1.14 Commercial sugar esters

1.5 Glycoside Surfactants

A glycoside consists of a sugar headgroup and an alkyl chain that is attached via the glycosidic bond at the anomeric carbon. Chemically it is an acetal, which does not exhibit any sensitivity towards bases. It is prepared by glycosylation of an alcohol. A glycolipid surfactant requires an alkyl chain with minimum 6-8 carbons to enable the surfactant activity.

1.5.1 Glycosylation strategy

There are several methods used for glycosylation. Probably the most common is called "Fischer glycosylation". In 1893, Emil Fischer (1893) came up with a direct synthesis of glycosides; an acid-catalyzed reaction of reducing sugars with alcohols (figure 1.15). Although this synthesis route successfully produces alkyl monoglucosides, a complex mixture of di-, tri- as well as oligoglycosides was also produced. Besides the dominating pyranosides and furanosides are also formed, which leads to complex product mixtures. Therefore isolation of individual molecular species is not easy (von Rybinski and Hill, 1998).

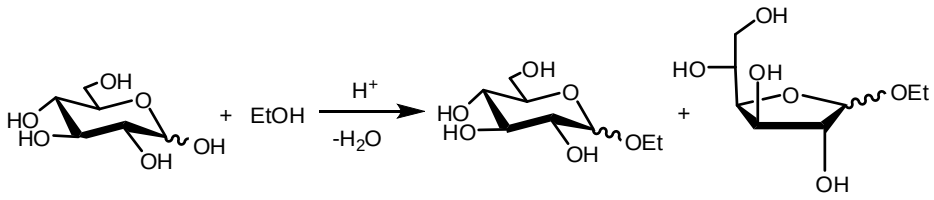


Figure 1.15 Fischer glycosylation

Later, Koenigs-Knorr (1901) proposed a novel glycosylation method which has been used most widely to form stereoselective glycosyl bonds (figure 1.16). The protecting group at the 2-position leads to 1,2-trans glycosides in the presence of silver or mercury promoters (Lindhorst, 2000). However there are two major disadvantages; the intrinsic lability of glycosyl halides and the need of Ag_2CO_3 or $Hg(CN)_2$ (Helferich and Weiss, 1956) as promoters which are inconvenient due to toxicity and price.

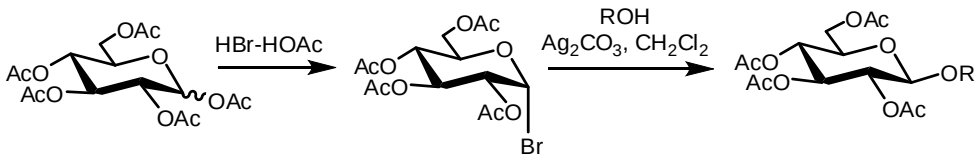


Figure 1.16 Koenig-Knorr reaction

Nowadays modern production plants are built based on the Fischer glycosylation to produce alkyl polyglycosides (APGs) in industrial scale. Over the years, development on the synthesis has been made to increase the efficiency in order to yield a better quality and economical product. The ratio of alkyl monoglycosides to alkyl polyglycosides can be control by adjusting the amount of glucose and alcohols. Relevant performances properties such as hydrophilicity can be adapted without further purification of the final product mixture or isolation of one component.

1.5.2 Alkyl polyglucosides (APG)

Alkyl polyglucosides, or APG, are based on Fischer's early work who synthesized a mixture of alkyl glucosides by glycosylation of ethyl alcohol with glucose. The characteristic of an APG depend on the alkyl chain and the degree of polymerisation (DP), which is the average number of linked glucose units.

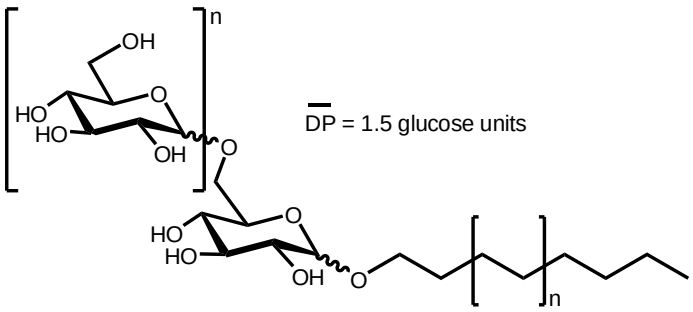


Figure 1.17 Alkyl polyglucosides (major structure)

The application of APG as surfactant in detergents began about 40 years after Fischer first discovered alkyl glucosides. Since then, efforts to produce APG at industrial scale have been performed by various companies. In late 1970's, Rohm & Haas were the first to market an octyl/decyl (C_8/C_{10}) polyglucosides. However short chain APGs gave unsatisfactory performance as surfactant due to the poor color quality that limited the

application as detergent (von Rybinski and Hill, 1998). Even though improvements have been made on the octyl/decyl (C_8/C_{10}) polyglycosides, longer chain APG (C_{12-14}) were produced in the late 1980's to meet demands for cosmetics and detergent applications (von Rybinski and Hill, 1998).

The synthesis of APG involves the combination of a reducing sugar (normally glucose) with alcohols. There are two ways to synthesis the APG, chemical and enzymatic synthesis. However only the chemical synthesis is applied for large scale production. The modern production plants are based on the Fisher glycosylation. This complies with the production criteria; i.e. to produce surfactants with suitable properties, economical and also minimise waste and emissions. Enzymatic synthesis would be an alternative method. Although an enzymatic synthesis could be a one step reaction process, high cost production or unavailability of suitable enzymes, make it impossible to be applied in industry. Moreover the synthesis is thermodynamically controlled. Besides the glycoside the reaction forms water. Because of this, high water concentration's shift the equilibrium towards the starting materials. However, the enzymes mostly required an aqueous medium. That makes the process thermodynamically disfavoured.

Effects of alkyl chain length and the degree of polymerisation have been studied to increase the understanding of APG behaviour (Fukuda et al., 1993; Nilsson et al., 1998; Bonicelli et al., 1998; Balzer et al., 1993). Most research concentrate on pure alkyl glycosides and only a few investigations target on APG. Applying pure AG as surfactant is too expensive, therefore APGs should be studied extensively in order to understand their properties. Von Rybinski and Hill (1998) have written an impressive review on properties and applications of APG.

1.5.3 Physicochemical properties

Interfacial properties of pure alkyl glycosides have been studied and compared with technical polyglucosides (APG). Shinoda et al. (1989) and Böcker et al. (1989) have determined CMC values of these compounds as a function of alkyl chain and the degree of polymerisation (DP). It is shown that the CMC is strongly influenced by the alkyl chain length whereas the number of glucose units affects the APG less. Furthermore, the CMC of pure alkyl monoglucosides and technical grade polyglucosides are comparable with other typical nonionic surfactants (i.e. alkyl PEGs), which decrease in CMC with increasing alkyl chain length.

Phase diagrams of various glycosides have been published over the years. There are many methods used to determine the phase diagrams for glycosides. Dörfler and Göpfert (1999) and Häntzschel et al. (1999) have investigated short chain (C_7-C_{10}) glucosides using DSC (differential scanning calorimetry) and the contact penetration method under an optical polarising microscope, while Zhang et al. (1999) used a combination of synchrotron X-ray and neutron scattering technique to analyse heptyl-, octyl- and nonyl β-glucoside. Time resolved fluorescence quenching was also used in comparing the micelle formation of octyl glucoside and dodecyl maltoside (Zana and Aoudia, 1998). Studies of the influence of stereochemistry, alkyl chain length and size of the sugar headgroups on phase transitions will be discussed further in subtopic 1.5.4.

1.5.4 Liquid crystal behaviour

Long chain glycosides like any other surfactants, exhibit liquid crystalline phases due to the amphiphilic structures that lead to self assembled aggregates when in contact with a solvent. However, liquid crystal phases, or in other words mesophases, may be observed

when pure glycosides are heated at high temperature. Therefore glycosides are known to be amphotropic liquid crystalline compounds.

There are three types of liquid crystals; thermotropic, lyotropic and amphotropic. For thermotropic liquid crystals, mesophases depend on the temperature only (heating/ cooling) whereas lyotropic liquid crystals form self-assemblies by contact with a solvent at certain concentrations, which also depend on the temperature. Thermotropic and lyotropic liquid crystalline behaviour can be exhibited by monophilic as well as by amphiphilic compounds. An amphotropic compound combines the thermotropic and the lyotropic behaviours.

Self-assembly behaviour of monophilic lyotropic and amphotropic liquid crystals differs significantly from each other. A monophile forms self-assemblies as a result of packing and/or interactions of extended π-systems, while an amphiphile shows mesophases because of the separation of two incompatible molecular regions, which tend to aggregate separately.

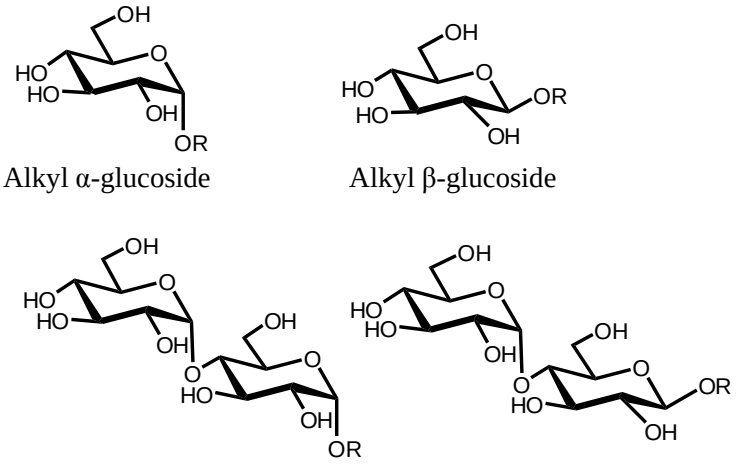
1.5.4.1 Thermotropic properties

Thermotropic behaviour of glycolipids was first observed in 1911 when Emil Fischer experienced a double melting point for hexadecyl glucopyranoside (Fischer and Helferich, 1911). Yet only in 1938, Noller and Hori (1938) described this behaviour as liquid crystalline and extended this phenomenon to other families of glycolipids.

Pure alkyl glycosides, which are major components in APG such as n-octyl β-glucopyranoside, n-decyl β-glucopyranoside, n-dodecyl β-glucopyranoside and their anomeric counterparts, have been studied extensively. Boyd et al. (2000) have investigated major components in APG looking at chain length, headgroup

polymerization and anomeric effects on their thermotropic and lyotropic liquid crystalline properties.

Most alkyl glucosides only exhibited smectic A phase upon heating (Jeffrey and Wingert, 1992). In the smectic A phase (figure 1.19) molecules tend to arrange in layers and pointing along the director. The director is perpendicular to the planes in which no positional order exists. By using OPM (optical polarising microscope), a unique texture can be observed resulting from the anisotropic character of the phase.



R = C_8H_{17}, $C_{10}H_{21}$, $C_{12}H_{25}$

Figure 1.18 Most studied alkyl glycosides

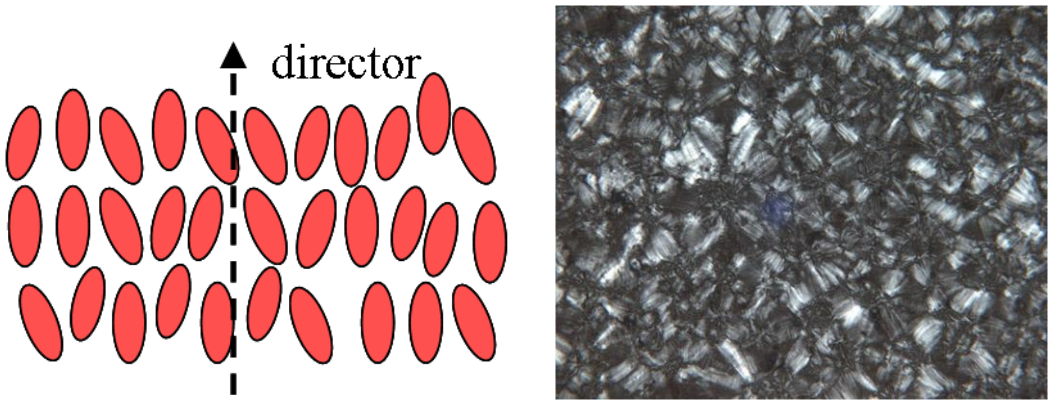


Figure 1.19 Arrangement in smectic A phase (left) and its corresponding texture observed under OPM (right)

Two types of transition temperature are observed for single chain glycosides; the melting temperature (crystal to smectic A) and the clearing temperature (smectic A to isotropic). These transition temperatures are influenced by several factors, such as headgroup size, alkyl chain length and anomeric configuration.

The melting temperature is influenced by the alkyl chain length whereas the clearing point mostly depend more on hydrogen bonding between the headgroups (Koeltzow and Urfer, 1984). The effect of the headgroup polymerization was observed by comparing n-dodecyl β-glucoside, n-dodecyl β-maltoside and n-dodecyl β-maltotrioside (Vill et al., 1989). The clearing temperature increased significantly with the size of the headgroup, i.e. from glucoside to the maltotrioside. However, the maltotrioside differs only slightly from the maltoside. Later von Minden et al. (2000) observed a 127K increase in the clearing temperature between n-stearyl β-glucoside and n-stearyl β-maltoside. It was explained that addition of hydroxyl groups lead to more hydrogen bonding in the headgroup region. The stability of smectic A phases for the glycosides was also enhanced with increasing sugar size, i.e. by replacing a monosaccharide with a disaccharide headgroup. Prada et al. (1995) reported that optimal hydrophilic and hydrophobic balance for monosaccharide headgroup is obtained with alkyl chains

containing 12-14 carbon atoms, whereas for disaccharide headgroup longer alkyl chain are needed to reach the optimal balance.

Higher clearing temperatures are observed with increasing alkyl chain length. However the introduction of unsaturation in the alkyl chain decreases the clearing temperature. This affect is larger for monosaccharides than for disaccharides. Studies of stearyl β-maltoside and oleyl β-maltoside reveal a difference of only 7K in clearing temperature (von Minden et al., 2000) whereas the corresponding glucosides show a 20K difference (Vill et al., 2000).

Hashim et al. (2006) investigated branched chain glycosides with a similar number of carbon atoms. While straight chain glycosides only exhibit one mesophase, branched chain glycosides can exhibit smectic A, columnar and cubic phases. Most of the branched chain glycosides form liquid crystalline phases at ambient temperature, except lactosides, which have high melting points.

The anomeric configuration between headgroup and alkyl chain also influences the liquid crystalline behaviour. For glucosides, α-anomers are thermally more stable than β-anomers (Sakya and Seddon, 1997). On the other hand, β-anomers are more stable than α-anomers for maltosides and lactosides (von Minden et al., 2000). This anomeric effect decreases with the chain length (Boyd et al., 1997).

1.5.4.2 Lyotropic properties

Lyotropic liquid crystalline phases are influenced by the same effects as thermotropic mesophase behaviour. Stubenrauch (2001) has summarized the factors that influence the thermotropic and lyotropic behaviour of glycolipids (short chains) in a review paper.

The headgroup polymerisation in APG has high impact on lyotropic mesophases. With increasing number of glucose, the water solubility of the material increases. Lack of OH groups on the sugar ring will lead to insufficient intermolecular hydrogen bonding, thus decreasing the solubility (Sakya and Seddon, 1997). Therefore, no phase separation was observed in maltosides compared to glucosides with the same chain length in partial binary surfactant-water (Boyd et al., 2000). The glucosides formed a phase separation because of high Krafft point. While alkyl β-glucosides form hexagonal (H_I), cubic (Q_I) and lamellar (L_α) phases with increasing surfactant concentration, alkyl β-maltoside showed H_1, anisotropic (H_I, L_α) phase and at higher concentration, L_α phase. The longer chain glucoside and maltoside also experienced the same behaviour (von Minden et al., 2000; Boyd et al., 2000).

Nilsson and co-workers (1998) stated that, when one carbon was added to the alkyl chain, the hexagonal phase melts at lower temperature (octyl β-glucoside and nonyl β-glucoside). However the hexagonal phase disappeared as the chain becomes longer and only lamellar phase was observed. Nevertheless maltosides showed similar behaviour even with increase chain length (Boyd et al., 2000).

When a stearyl chain was replaced with an oleyl chain, a broad bicontinuous cubic V phase was exhibited along with lamellar, hexagonal and anisotropic phases. The oleyl β-maltoside displayed in sequential order; lamellar (L_α), cubic (V_1), hexagonal (H_1) and anisotropic phases (von Minden et al., 2000). Yet for glucosides, only myelin figures were formed when contacted with water due to very low water solubility (Vill et al., 2000).

The anomeric configuration also affects the lyotropic phases. This effect strongly influences the lyotropic phases for shorter alkyl glycosides; however for longer alkyl

chain glycosides, it becomes weaker. Nilsson et al. (1998) reported the difference in mesophases observed on octyl α-glucosides and octyl β-glucosides. Octyl α-glucosides exhibited phase transitions of micellar, hexagonal and lamellar as the surfactant concentration increased. Conversely in the case of octyl β-glucosides, there are micellar, cubic (bicontinuous) and lamellar phase present. Furthermore the Krafft boundary for α-glucosides was observed to be higher than for β-glucosides (Dorset and Rosenbusch, 1981) due to stable crystal packing of the α-anomer in pure state. Two branched chains compounds, C_2C_6-α-glucoside and C_2C_6-β-glucoside are also reported in the same paper. They only showed two phases; micellar and lamellar. Yet these phases behaved differently with respect to the concentration range of the two phase region (observed from the phase diagram) for the β-anomer decreased with increasing temperature while it is remained unchanged for the α-anomer.

1.6 Objectives of the Study

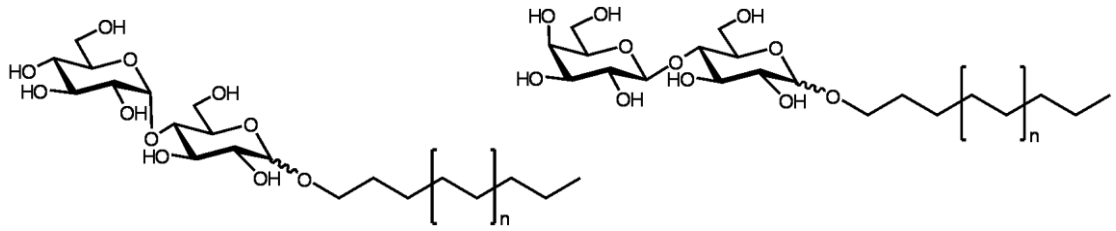
APG has been applied as non-ionic surfactant in detergent and agricultural industries for many years. It is also used as emulsifier in cosmetics due to high skin compatibility. APG is made from natural resources which are biodegradable and harmless to the environment, thus making it as one of the most important surfactant nowadays. However, due to the complex chemical compositions, APG is not suitable for pharmaceutical or other bio-related applications as biological tests are required for each component. Moreover, having an average polymerisation degree of the sugar head of ~1.5, APG is lack on hydrophilic domain. This limits the application as emulsifier in water based systems (oil in water).

The objective of this research is to synthesize technical grade glycosides similar to APG from palm oil and palm kernel oil. These glycosides comprise of disaccharide

headgroups, which increase the hydrophilic behaviour and, thus, increases the emulsifying properties. The sugar headgroup is based on maltose and lactose considering economical aspects. Palm oil and palm kernel oil are used as the source of fatty alcohol in order to promote local resources. The synthesis method was adapted from Vill et al. (1989), to produce "clean" technical glycosides comprising only of different anomers and, in case of palm (kernel) oil based products, including alkyl chain variations. Thus these glycosides will be suitable for applications in pharmaceutics and cosmetics products.

A range of glycosides was synthesised and their liquid crystal properties were investigated, i.e. thermotropic and lyotropic as well as interfacial properties for selected compounds. The study of palm oil and palm kernel oil glycosides involves the effects of stereoisomers and chain inhomogeneity. Model studies have been perfomed in order to understand each effect. Various α/β mixtures of dodecyl maltosides and lactosides were prepared to investigate the stereochemical effect, whereas the chain inhomogeneity effect was studied by comparison of dodecyl (C_{12}), cetyl (C_{16}) and oleyl ($C_{18:1}$) chains. The selection of chains is based on major components in palm oil and palm kernel oil. The interfacial properties of dodecyl maltosides and lactosides were also determined to study the stereochemical influence. Both effects have been discussed in topic 1.5 for pure glycosides.

The general structures for both alkyl maltosides and alkyl lactosides that have been synthesised are shown in figure1.20.



(a) (b)

Figure 1.20 General chemical structures of (a) alkyl maltoside and (b) alkyl lactoside

2.1 Synthesis

The glycolipids were synthesised using lactose or maltose as sugar headgroups whereas alcohol mixtures as the lipid tail. The overall synthesis method consist three major steps, i.e. protection and activation of the sugars (acetylation), glycosylation and deprotection of the sugars (deacetylation). The alcohol mixtures were produced from the reduction of palm oil and palm kernel oil. Figure 2.1 shows the overview of the synthetic route.

2.1.1 Preparation of palm oil based alcohols

Palm oil and palm kernel oil were reduced to alcohol mixtures using the method referred from Nystrom et al. (1947). Lithium aluminium hydride is a reactive reducing agent which can reduce carboxylic acids and esters to the corresponding alcohols effectively. Moreover double bonds are not affected during the reduction. This is an advantage because both palm oil and palm kernel oil contain double bonds in significant amounts.

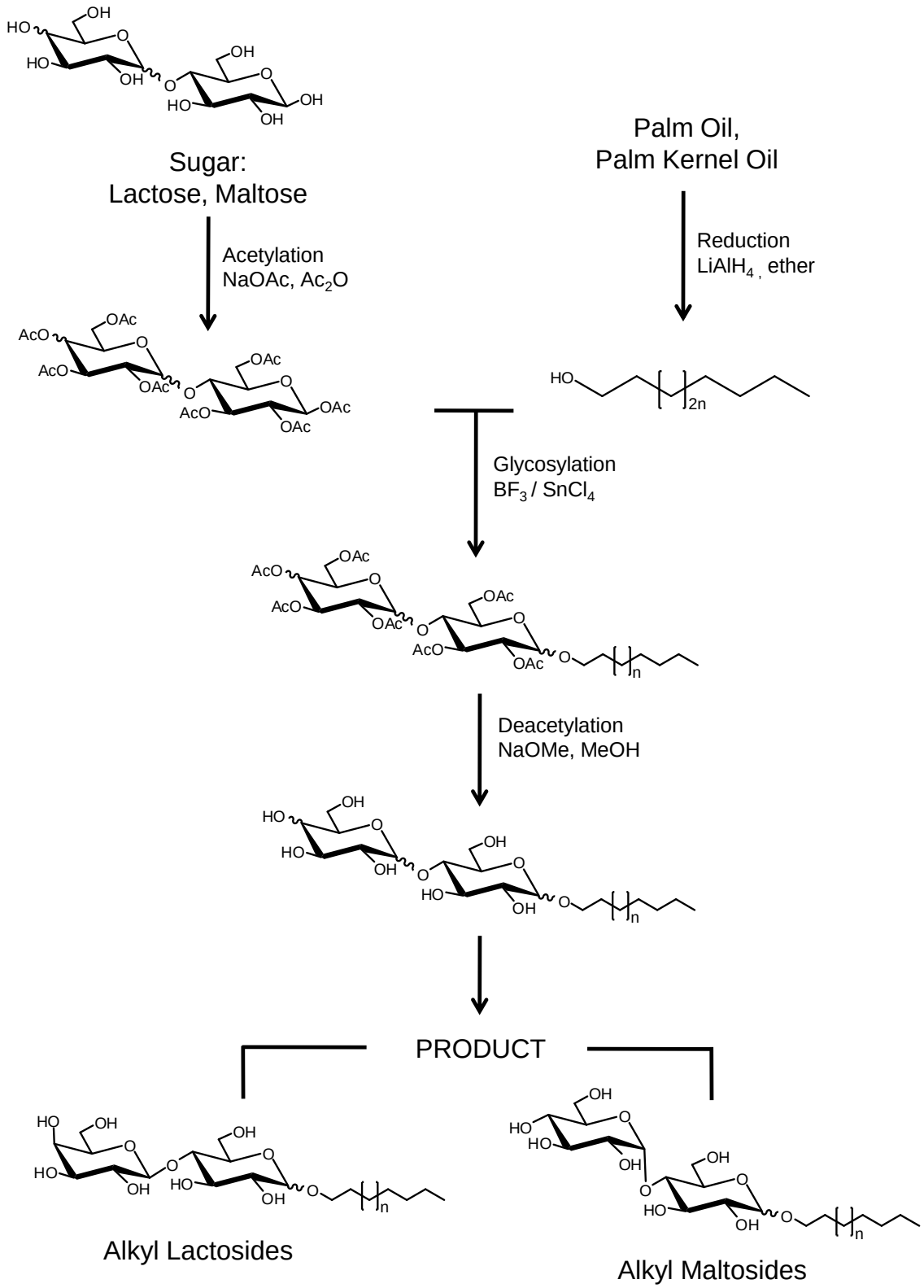


Figure 2.1 Synthetic route overview

2.1.2 Activation of sugars

In order to have a selective reaction at the anomeric centre, hydroxyl groups of the sugar must be protected. Suitable protecting groups ensure high yields for the introduction and stability towards glycosylation. Besides, an activation of the anomeric center would be beneficial. It is also useful in terms of solubility, since the sugar and a fatty alcohol are usually immiscible. The criteria for the selection of the ideal protecting group are as follows:

- efficiency – easy introduction of the group;
- stability – not affected during reaction;
- ease of removal (deprotection step);
- costs.

Based on these criteria, acetate is the most suitable protecting group in our case.

There are a few synthesis methods available for acetylation. Mostly are using acetic anhydride. An equal proportion of acetic anhydride and pyridine produces an anomeric mixture containing both α- and β-anomers in a ratio depending on the starting material (Haworth et al., 1937). Acetylation using acetic anhydride catalysed by a Lewis acid, i.e. zinc chloride, has been reported for α-dominant mixtures (Malm et al., 1953). Since β-glycoside is most suitable for glycosylation, acetylation with acetic anhydride and sodium acetate was chosen as these conditions provide β-glycosides as major products (Vogel, 1989; Vill et al., 1989).

2.1.3 Glycosylation

For the production of technical grade alkyl lactosides and alkyl maltosides, an approach following Vill et al. (1989) and. von Minden et al. (2000) was adapted. A slightly modified method has also been applied for the production of pure branched chain glycolipids by Hashim et al. (2006). Either tin tetrachloride ($SnCl_4$) or boron trifluoride (BF_3) was used as catalyst to produce mixtures varying in the anomeric ratio (figure 2.2).

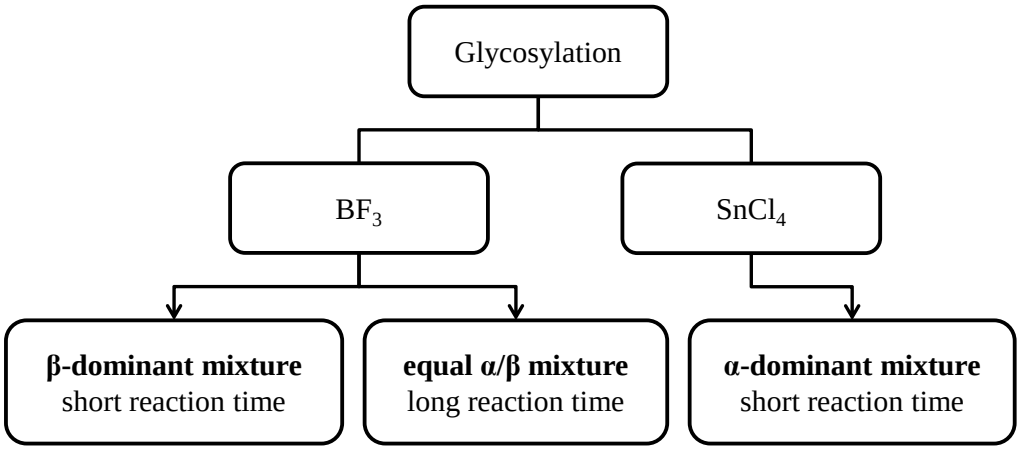


Figure 2.2 Glycosylation flowchart summarizing the product synthesis

The ratio of α- and β-anomers depends on parameters such as reaction time and catalyst because β glycosides are kinetically favoured, whereas α glycosides are thermodynamically more stable.

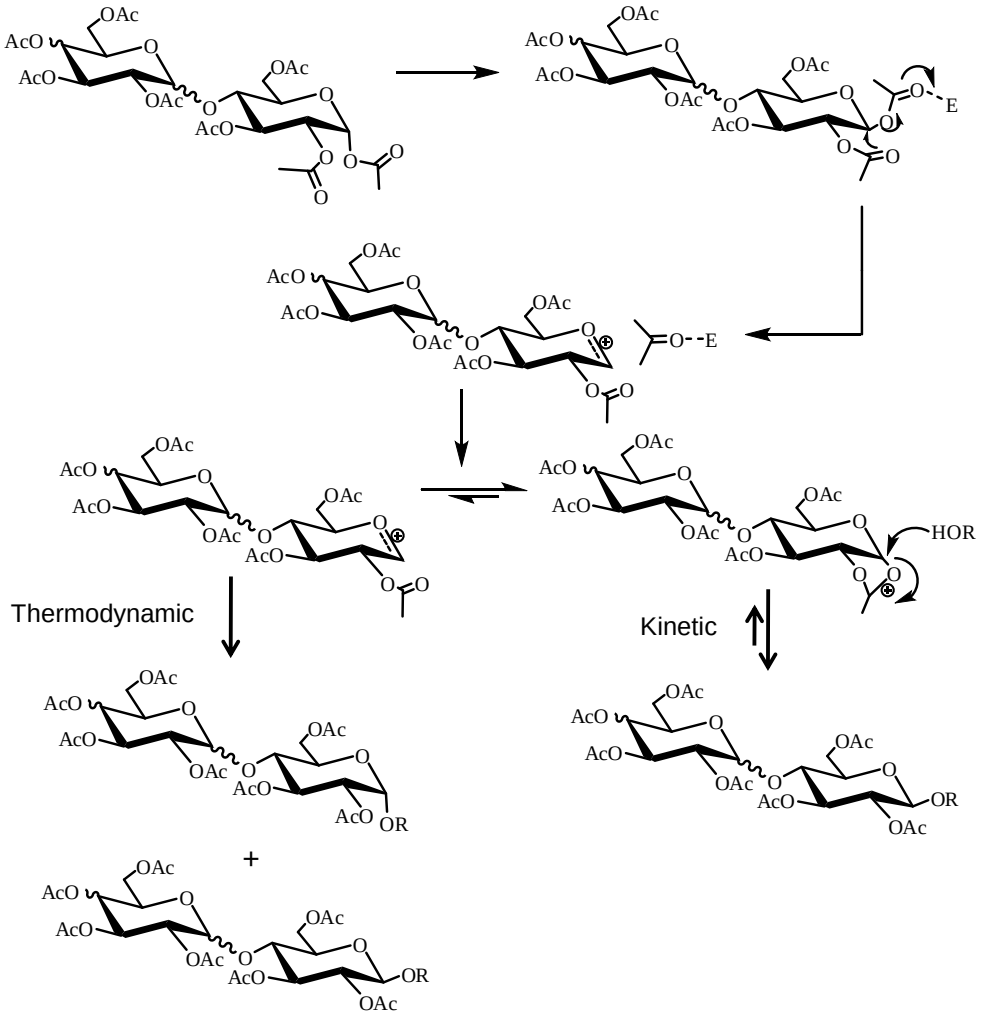


Figure 2.3 Reaction mechanism

The carbonyl oxygen from the neighbour group (acetate at O-2) stabilizes the intermediate glycosyl cation and forms a cyclic oxonium ion (figure 2.3). This shields the α position, thus favouring the β-anomer. In order to produce the α-anomer, which is thermodynamically preferred, high activation energy is needed to be able to form back the cyclic oxonium ion because at this point the glycosidic oxygen has low electron density. The formation of α-anomer is due to the anomeric effect as shown in figure 2.4. The dipole moments of α-anomer are partially neutralizing each other whereas they increase for β configuration (Lindhorst, 1999). A stronger catalyst such as $SnCl_4$ is used

to produce α-dominant mixture. However reaction catalysed by BF_3 will also produce the same product if longer time is applied.

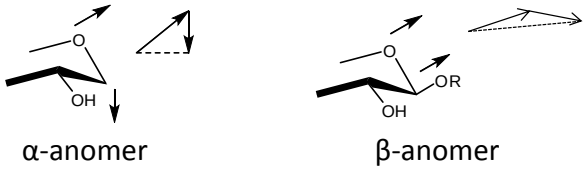


Figure 2.4 Anomeric effect

Only moderate precaution is required, i.e. reactions were operated in closed apparatus to prevent excess moisture. The complicated purification step is omitted since we are producing technical products. Excess alcohol from the product can be removed by a simple extraction process using acetonitrile and hexane.

2.1.4 Deprotection of surfactants

The method used is the Zémplen deacetylation (Zémplen, 1929). The acetates were removed by treatment with sodium methoxide in methanol. Then diluted hydrochloric acid was used to neutralize the basic solution during an extraction process. Separation of glycolipid and remaining sugar was achieved through extraction with butanol and water.

2.2 Chemical Product Analysis

Products were analysed using the nuclear magnetic resonance (NMR) and iodine titration.

2.2.1 Nuclear magnetic resonance (NMR) spectroscopy

Acetylated glycolipids were analysed in deutro-chloroform while the final products were dissolved in methanol-d_4. The α-dominant dodecyl lactosides had to be dissolved in pyridine-d_5 due to their low solubility in methanol-d_4. Measurements were conducted at room temperature except for alkyl lactosides samples, which required a temperature of ~55°C due to insufficient solubility at lower temperature. All proton spectra were measured on a JEOL NMR spectrometer at 400 MHz.

Analyses of NMR spectra enable the evaluation of the product quality and also indicate the anomeric ratio of the mixture. This can be obtained by comparing the integral of the α-anomer and the β-anomer peak. A proper baseline and phase correction is required to avoid systematic errors in determining the values. The quality of the product was determined from the ratio of sugar and lipid chain. In the synthesis excess alcohol is used and then removed by extraction. However improper extraction will not remove the alcohol completely thus leading to remains in the product. Unreacted alcohol is not easily detected in NMR spectrum since the signals of the alcohol CH_2 overlap with sugar signals. Therefore the ratio of sugar and lipid chain is used to determine the presence of this alcohol. The average length of alkyl chains in products from palm oil and palm kernel oil can also be calculated from their peak integrals. Figure 2.5 shows the NMR spectrum of a dodecyl maltoside with important peaks to determine the

characteristic of the product. Figure 2.6 shows the anomeric ratio determination based on peak integrals.

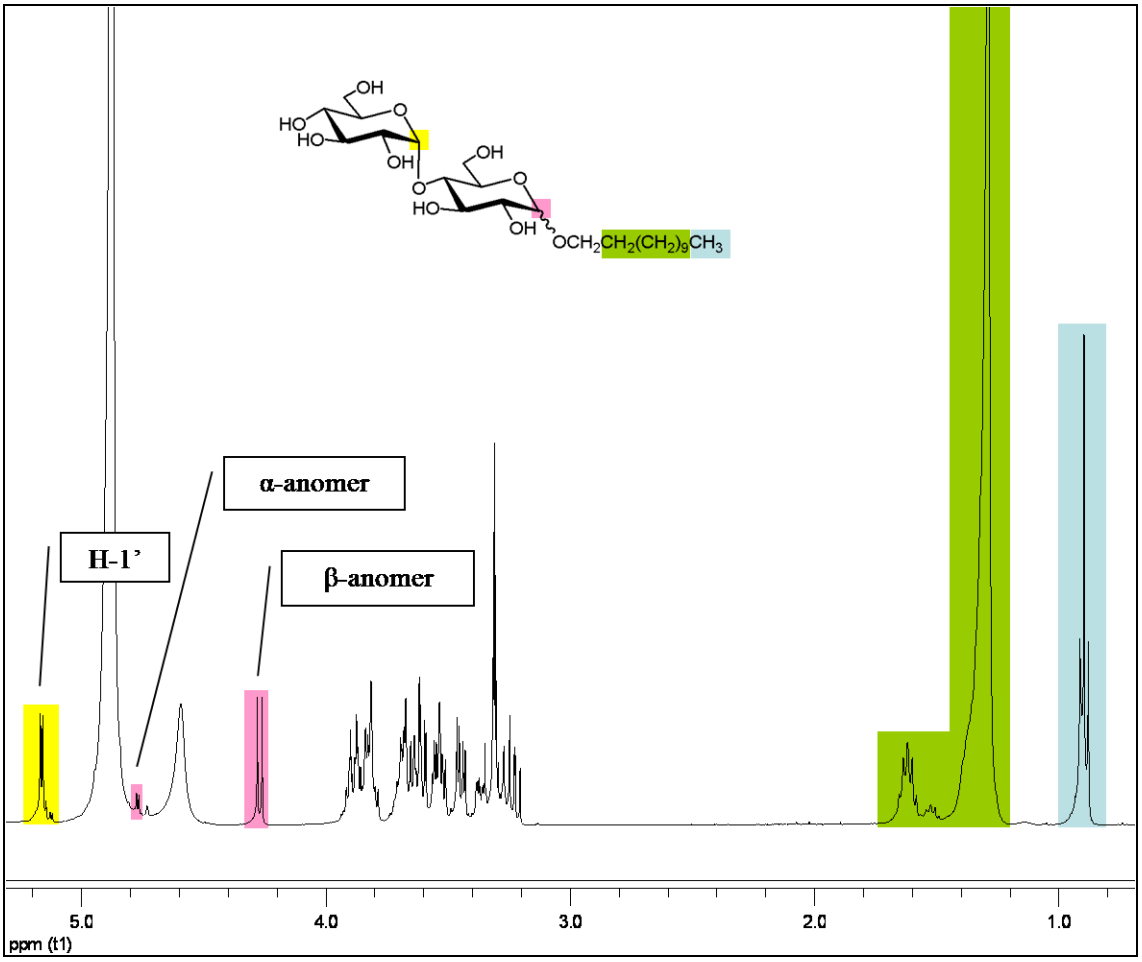


Figure 2.5 ^{1}H-NMR spectrum of dodecyl maltoside

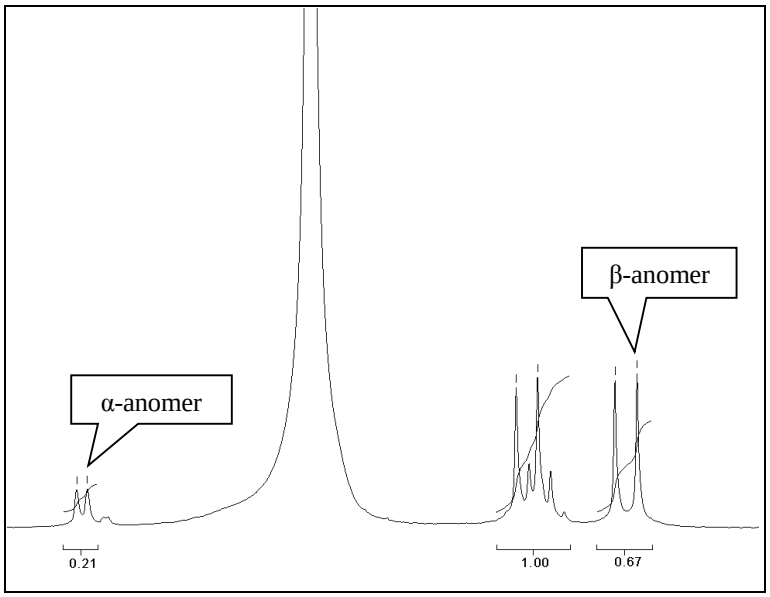


Figure 2.6 Example of α/ β ratio determination from peak integrals for a lactoside

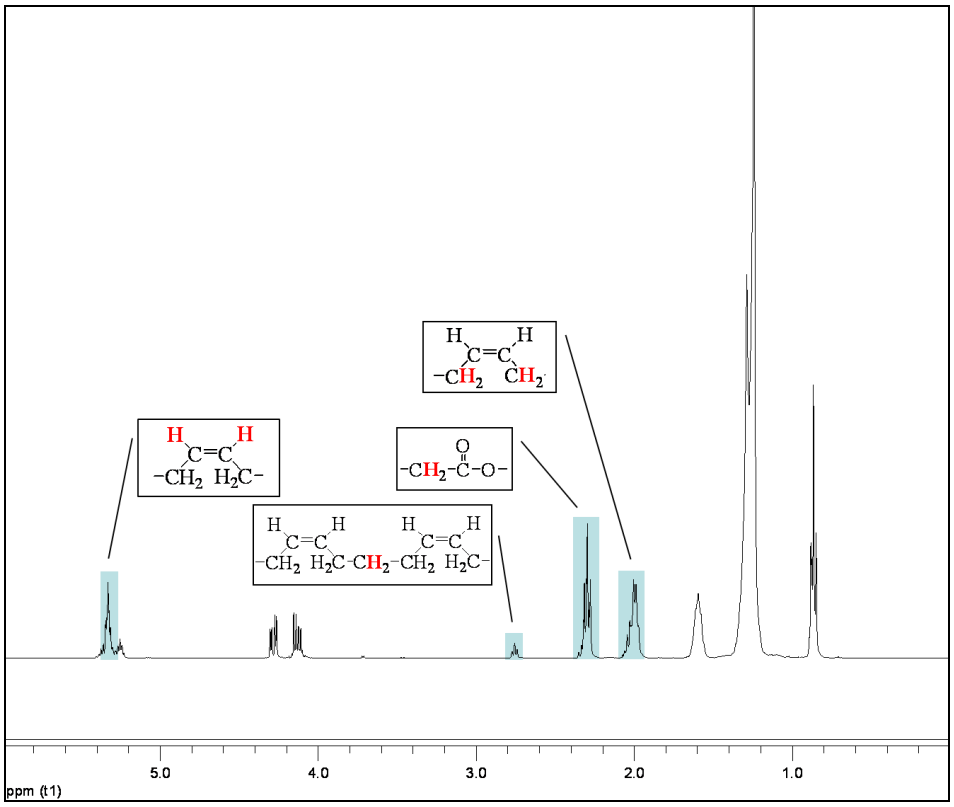


Figure 2.7(a) ^{1}H-NMR spectrum of palm oil

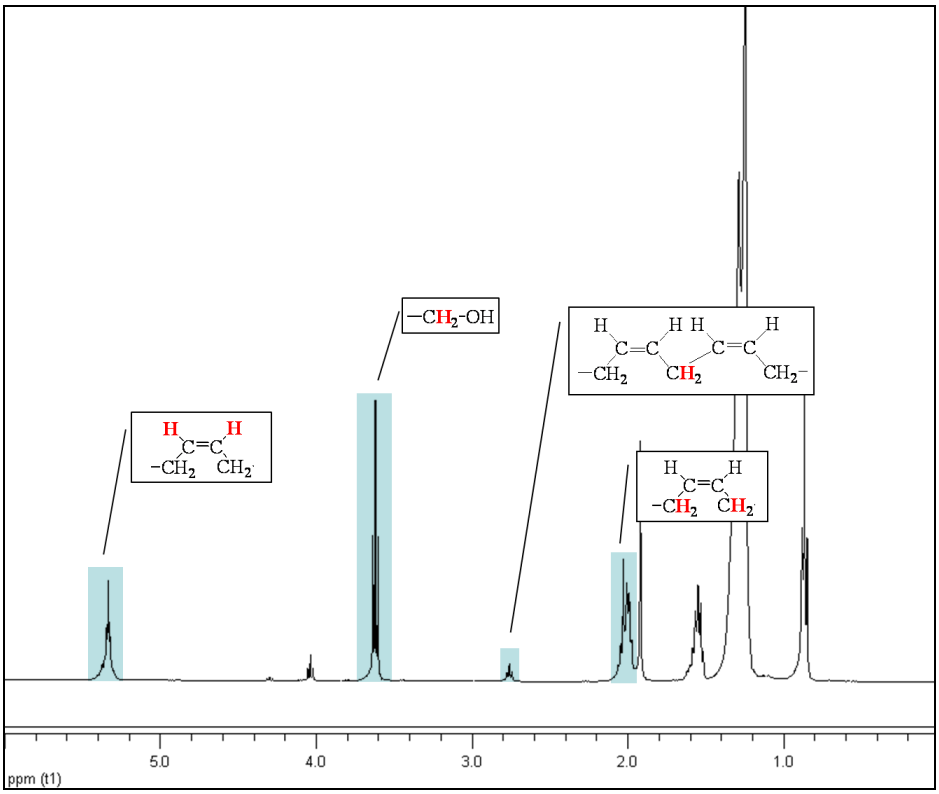


Figure 2.7(b) ^{1}H-NMR spectrum of reduced palm oil

The spectrum for palm oil reduction is shown in figure 2.7. The product showed presence of alcohol peak at 3.8 ppm thus confirming the conversion of esters to alcohols, while the absence of the ester peak (~2.2 ppm) indicates no leftover starting material.

From the spectra, the unsaturation content can be roughly estimated based on the integration of the peaks. This applies to palm oil and palm kernel oil, as well as their respective glycolipids. The overall content of unsaturation can be calculated from integration of the peak at 5.3 ppm whereas the polyunsaturated compound can be estimated from the peak at 2.7 ppm. The content of monounsaturation was calculated by subtracting the polyunsaturation content from the peak at 2.0 ppm. The results were checked by iodine titration. The unsaturations in glycosides were determined in the same way.

2.2.2 Iodine titration

The degree of unsaturation for palm oil and palm kernel oil was also determined by iodine titration using Wijs solution. This method is according to the American Society for Testing and Materials (ASTM). The experiment involves the additions of iodine to the double bonds. The excess reagent is titrated and compared to a blank. Wijs solution is commercially available and was used directly; however, due to degradation of the reagent the titer was checked before use.

2.3 Physical Analysis - Liquid Crystal Investigation

As mentioned in 1.5.4, the glycosides exhibit both thermotropic and lyotropic liquid crystal behaviours. Different methods were used to analyse these behaviours.

2.3.1 Thermotropic behaviour

Thermotropic behaviour of a liquid crystal compound depends on the temperature. A pure liquid crystal material exhibits phase transitions as the temperature change.

2.3.1.1 Optical polarising microscopy (OPM)

Liquid crystal phases can be identified by characteristic textures while phase transitions can be observed by changes in the texture under an optical polarising microscope. Optical polarising microscopy is a method used to study optically anisotropic compounds.

An image is produced as a result from the interaction of linear polarised light with a birefringent compound. The electric field vector of polarised light oscillates in a certain

direction, hence a linear polarised light oscillates in a plane. Normal light travels in many directions at different wavelengths therefore it is called non-polarised light. In order to have a polarised light, polarisers are used to separate and eliminate other light and leave only a specific direction, or polarised light. When the direction of the light is parallel to the orientation of the polariser, the light can pass. On the contrary, if the light's vector is perpendicular to the polariser, it will be blocked and cannot pass. Light in an intermediate position, which is between parallel and perpendicular, will be absorbed partially. However if two polarisers are placed perpendicular to each other, no light can pass through.

The optical polarising microscope applied the light polarisation principle. It is equipped with 2 polarisers, the first polariser is placed in the light pathway under the sample holder and the second polariser or the analyser is situated in the optical pathway between objective rear aperture and the observation tubes or a camera port.

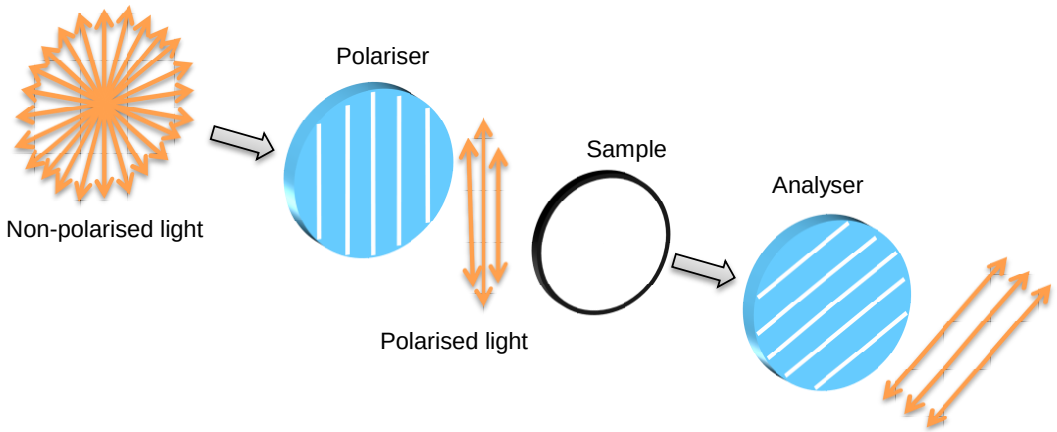


Figure 2.8 Principle of light polarisation of OPM

In case of a liquid crystal, a texture is observed due to the birefringence nature of the material. This happens when the polarised light that passes through the material split into two orthogonal beams that travel at different speed. They are called ordinary ray

and extraordinary ray. The light components are recombined by the analyser based on their constructive and destructive interference. The instrument used in the investigation is an Olympus BX52 optical polarising microscope equipped with a Mettler FP82 Hotstage (heating stage).

A thin layer of sample was placed sandwiched between slip cover and the glass slide. The sample was heated from 30°C to 250-300°C. The end temperature depends on the clearing point of the sample, therefore every sample has a different end temperature. Phase transition temperatures were recorded at a heating rate of 5°C/min. Since the compounds investigated are mixtures, all temperatures are reported in a certain range instead of one specific value. Images of the texture were captured and stored using the analysis® Imager software by Soft Imaging System GmbH. Mesophases exhibited by samples were identified based on the texture. The melting and clearing temperatures were confirmed by differential scanning calorimetry (DSC).

2.3.1.2 Differential Scanning Calorimetry (DSC)

Differential scanning calorimetry (DSC) is a common thermoanalytical technique to measure melting and clearing points for liquid crystalline materials. This technique measures the differences in the heat flow between the sample and a reference (Skoog et al., 1998). Sample and reference are maintained at nearly the same temperature throughout the experiments. Therefore when the sample experiences a phase transition, heat is absorbed or released in order to maintain the same temperature in both medium. A DSC thermogram exhibits positive and negative peak and is a display of the heat flow versus time. The enthalpies of transitions can be calculated from these curves.

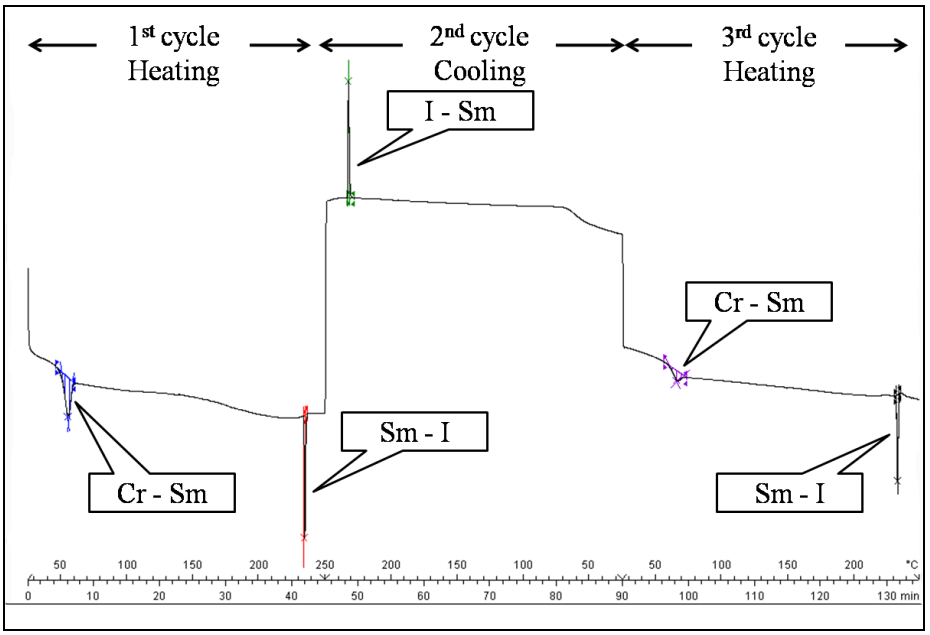


Figure 2.9 DSC thermogram

A Mettler Toledo DSC822[e] with Haake EK90/MT intra-cooler was used for the differential scanning calorimetry measurements. The data was analysed using STAR[e] Thermal Analysis System software.

Samples were measured based on a heating-cooling-heating cycle (figure 2.9). The temperature range applied was based on data obtained from OPM studies. In order to observe the reformation of mesophases, samples were first heated to isotropic state, then cooled down to the initial temperature (30°C) and finally heated once more. A heating rate of 5°C/min was applied.

Samples, weighing 4-8 mg, were placed in standard aluminum pans of 40µl size. They were dried in a vacuum oven overnight and sealed with pinhole cover immediately before measurement.

2.3.2 Lyotropic behaviour

Lyotropic mesophases occur by contact with a solvent. The lyotropic behaviour depends not only on the concentration but also on the temperature.

2.3.2.1 Contact penetration

The method described by Vill et al. (2000) was applied in the investigation. The sample was placed on the microscope slide in a small amount and was pressed under a cover slide. In order to get a uniform texture for contact penetration, the sample was heated until its melting point then cooled to room temperature. Then solvent was introduced at the edge of covered slide. The solvent slowly moved towards the sample in a capillary action surrounding the sample before any contact occurred. The lyotropic phase was observed by OPM and identified based on texture. Water was used as the solvent in this investigation due to the application task of the material as surfactant. All experiments were performed at room temperature.

2.3.2.2 Critical micelle concentration (CMC)

The critical micelle concentration, or CMC, is an important characterisation of a surfactant. A surfactant, comprising of a hydrophilic head and hydrophobic lipid tail, tend to form aggregates as the concentration of the solution increases. These aggregates are called micelles. The concentration where micelles begin to form is known as the CMC. A good surfactant required a low CMC, thus making it more effective at low concentration. Most nonionics have low CMCs (Holmberg, 2003).

Surface tension measurement was applied to determine the CMC. The method was chosen due to the availability of the instrument. Applying the Du Nuoy ring method,

surface tensions of the samples were measured using KSV 702 tensiometer. Sample solutions were prepared with deionised water and were not measured directly after preparations (due to instrument accessibility). However the samples proved to be stable based on repeated measurements with varying storage times (1-30 days). All measurements were performed at 30°C.

A graph of the surface tension versus the $\log_{10}$ of the concentration (figure 2.12) was plotted for each sample. The CMC was determined as the point of intersection between two linear regression lines, i.e. the high concentration level off and the slope at lower concentration. The same method to determine CMC has been applied for pure β-dodecyl maltoside before (Niraula et al., 2004).

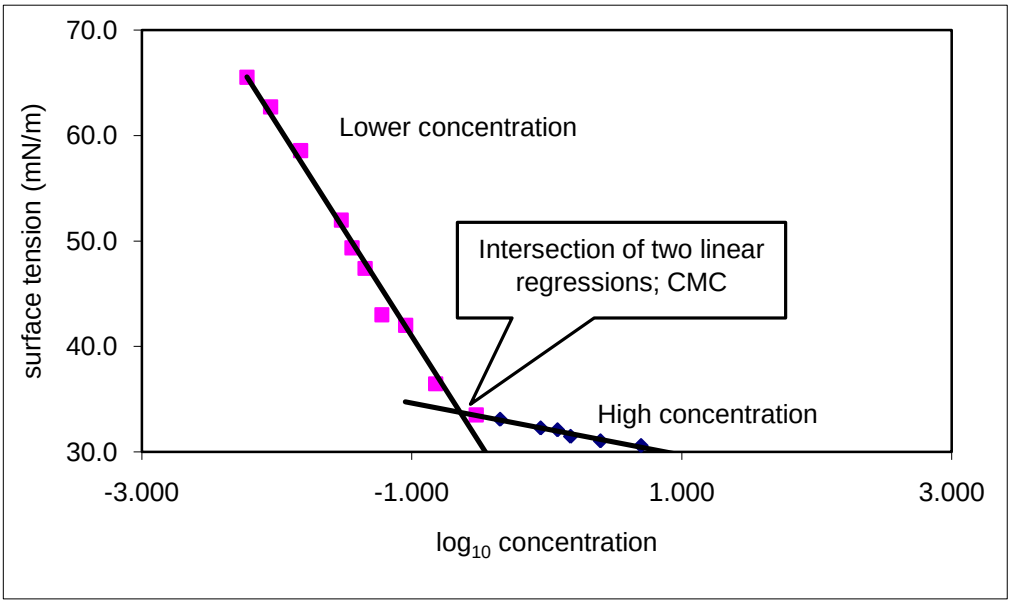


Figure 2.10 Graph surface tension versus $\log_{10}$ concentration for CMC determination

3.1 Materials

3.1.1 Alkyl glycosides

Besides maltoside and lactoside based palm oil and palm kernel oil, several glycosides with homogenous chain were synthesized. The purpose of these compounds was to enable a separate study of different influencing effects, such as anomeric and chain length effect. These are the major sources of product inhomogeneity for palm and palm kernel oil based glycosides. The synthesis route of the glycosides, as described in chapter 2.1, involved three major steps; acetylation, glycosylation and deacetylation.

3.1.1.1 Anomeric ratio

Variations of the glycosylation conditions led to mixtures with different α/β ratio. As mentioned before, the β-anomer is kinetically favourable. Therefore a short glycosylation time and a moderately active catalyst, boron trifluoride (BF_3), were applied to produce more β-anomer than α-anomer. On the other hand, a stronger catalyst, tin tetrachloride ($SnCl_4$), was used to produce α-dominant products, which are thermodynamically more stable. However α-dominant products can also be obtained from a BF_3 catalysed reaction, if longer reaction time is applied. The anomeric ratio of the products was determined from ^{1}H-NMR spectra as discussed in chapter 2.

Glycosylation with BF_3 is more preferable since the catalyst is easier to handle compared to $SnCl_4$. This is due to the formation of SnO_2 which complicates the reaction workup. Thus BF_3 was initially used to synthesize different anomeric mixtures. Six to seven hours reaction with BF_3 gave satisfying yields of 40-70% β-dominant products. With increasing time more α-anomer is formed. The reaction was monitored by thin layer chromatography (TLC). After 24 hours, a balanced ratio of α- and β-anomer was

obtained. Longer reaction times, i.e. 48 hours, produced α-dominant mixture with α/β ratios of ~2:1. Although the time can be increased further, it is no longer practical due to economical considerations. Therefore the catalyst was changed to $SnCl_4$ and a significant dominance of α-anomer was obtained.

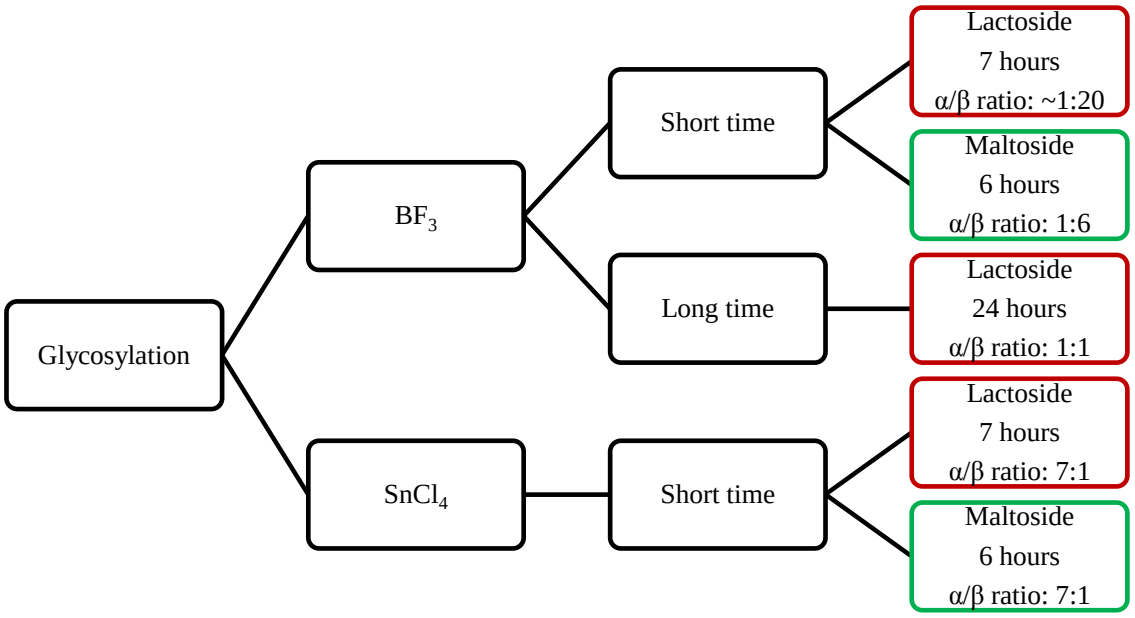


Figure 3.1 Synthesis of different dodecyl glycosides mixtures varying in α/β ratio

Figure 3.1 shows synthesis procedures for different anomeric mixtures for dodecyl lactoside and maltoside. Lactoside produced significantly more β-anomer than maltoside given the same catalyst and reaction time. However, this trend did not apply for α-dominant mixtures.

Increasing chain length, C_{16} instead of C_{12}, did not change the α/β ratio in case of the lactoside if the same reaction conditions were applied. However a slight increase in β-anomer ratio was obtained for hexadecyl maltoside,that is 1:6 instead of 1:5 for

dodecyl maltoside. The difference is insignificant within the error of determining the ratio.

In the case of BF_3 catalysed reactions, an increase of β-product selectivity was obtained for both oleyl maltoside and oleyl lactoside compared to their respective dodecyl glycosides. This corresponds to lower α-product selectivity for $SnCl_4$ catalysed reactions. Here only a 2:1 α/ β ratio of oleyl lactoside was obtained compared to the 7:1 mixture obtained for dodecyl lactoside under the same conditions. However the $SnCl_4$ reduced ~50% of the double bond content, whereas with BF_3 it was not affected.

Based on the synthesis of model compounds, the anomeric ratio of both lactosides and maltosides is not significantly affected by the alkyl chain length. The palm oil based lactoside and maltoside gave similar anomeric ratio to their respective oleyl counterparts based on the same synthesis parameters. Unlike for BF_3 catalysed reactions, $SnCl_4$ reduced the unsaturation content in palm oil glycosides significantly. The same was observed for oleyl lactoside. This clearly indicates a chemical reaction of the double bond with $SnCl_4$.

Despite its unsaturation content, palm kernel oil behaves similar than saturated alkyl chains. A twenty four hours reaction with BF_3 produced 1:1 α/β ratio palm kernel oil lactoside. A slight β-dominant (1:2) mixture was obtained for maltoside with six hours reaction catalysed by BF_3. Figure 3.2 shows the summary of the glycosides anomeric ratio based on synthesis condition. Although several different mixtures were synthesized, only the ones with comparable anomeric ratio were used for physical characterisations. Table 3.1 shows the α/β ratio of selected compounds determined by NMR spectra analysis. The purity of the mixtures was around 70-90% due to a small

quantity of a by-product that showed an additional H-1 peak for both maltosides and lactosides. This may indicate the presence of a seven-membered ring glycoside.

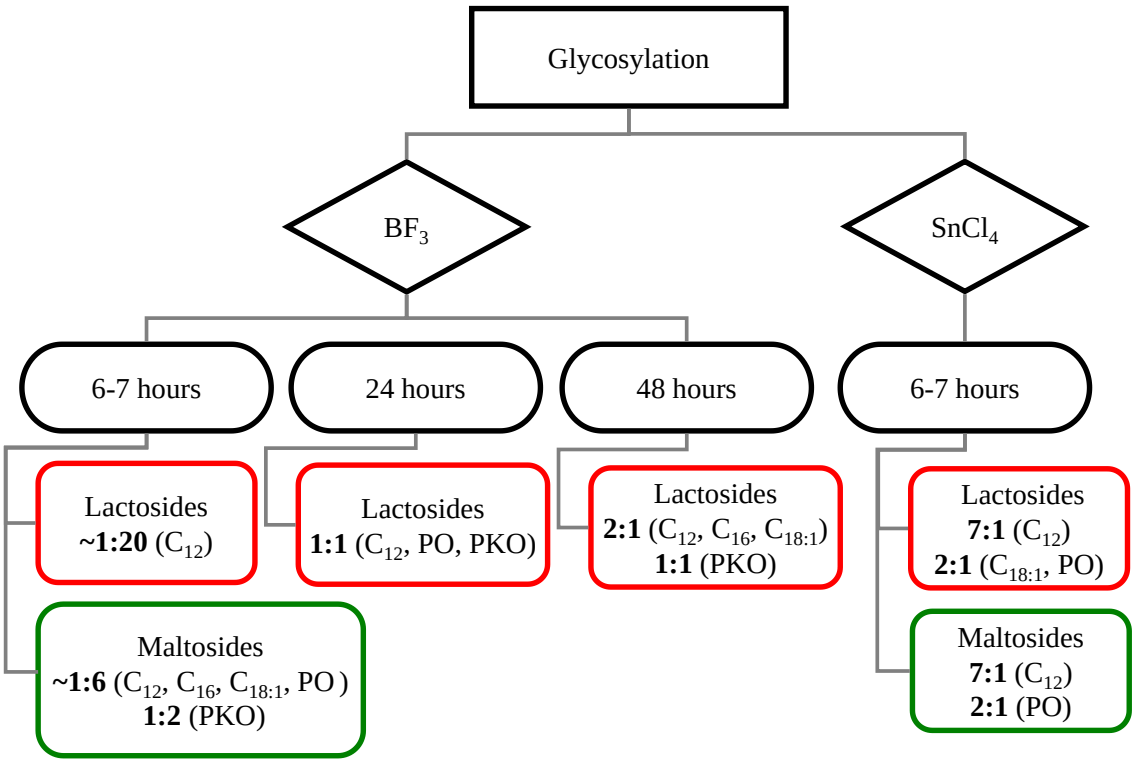


Figure 3.2 Summary of synthesis condition of lactosides and maltosides

Table 3.1 The α/β ratio of lactosides and maltosides varying in alkyl chain length

Sugar head	Alkyl Chain		α/ β ratio
Lactoside	Dodecyl ($C_{12}H_{25}$)	α-dominant	7:1
		balanced	1:1
		β-dominant	~1:20
	Hexadecyl ($C_{16}H_{33}$)		2:1
	Oleyl ($C_{18}H_{36}$) (BF_3)		1:1
	Oleyl ($C_{18}H_{36}$) ($SnCl_4$)		2:1
	Palm oil (BF_3)		1:1
	Palm oil ($SnCl_4$)		2:1
	Palm kernel oil (PKO)		1:1
Maltoside	Dodecyl ($C_{12}H_{25}$)	α-dominant	7:1
		β-dominant	1:5
	Hexadecyl ($C_{16}H_{33}$)		1:6
	Oleyl ($C_{18}H_{36}$)		1:6
	Palm oil (BF_3)		1:6
	Palm oil ($SnCl_4$)		2:1
	Palm kernel oil (PKO)		1:2

3.1.1.2 Reaction yield

Initially all compounds were synthesized in 2.7 mmol sugar scale. The yield ranged between 15 and 40%. However, the dodecyl glycosides were synthesized in larger scale, because they were needed for the determining of CMCs, which requires a lot of material. Therefore, glycosylation was performed based on 15 mmol scale and a satisfactory yield of >50% were obtained.

Table 3.2 Synthesis yields of various lactosides and maltosides

Sugar head	Alkyl Chain		Catalyst	Reaction scale (mmol)	Yield (%)
Lactoside	Dodecyl ($C_{12}H_{25}$)	α-dominant	$SnCl_4$	15	51
		balanced	BF_3	15	54
		β-dominant	BF_3	15	40
	Hexadecyl ($C_{16}H_{33}$)		BF_3	2.7	17
	Oleyl ($C_{18}H_{36}$)		BF_3	1.7	36
	Oleyl ($C_{18}H_{36}$)		$SnCl_4$	2.7	16
	Palm oil		BF_3	1.7	23
	Palm oil		$SnCl_4$	15	25
	Palm kernel oil (PKO)		BF_3	2.7	35
Maltoside	Dodecyl ($C_{12}H_{25}$)	α-dominant	$SnCl_4$	24	60
		β-dominant	BF_3	15	69
	Hexadecyl ($C_{16}H_{33}$)		BF_3	2.7	32
	Oleyl ($C_{18}H_{36}$)		BF_3	2.7	22
	Palm oil		BF_3	1.7	31
	Palm oil		$SnCl_4$	15	28
	Palm kernel oil (PKO)		BF_3	2.7	33

Palm oil based glycosides, on the contrary, were only obtained in 20-40% yield regardless of the reaction scale. This corresponds with lower yields for oleyl glycosides compared to other glycosides. The catalyst did not affect the yield for the saturated compounds. However $SnCl_4$ reduced the unsaturated compound thus lowers the yield. Table 3.2 reported the yield for the selected compounds.

3.1.2 Hydrocarbon chain analysis

Nuclear Magnetic Resonance Spectroscopy was used to analyse all the synthesised compounds, both on stage of peracetylated intermediate and the deacetylated final product. In this case, the deacetylated compounds spectra provided better information compared to the peracetylated compounds. This is because most of the important peaks are overlapped, thus it is difficult to analyse.

From the NMR spectra, the average alkyl chain length for reduced palm oil was seventeen carbons whereas palm kernel oil was almost fourteen carbons. Similar results were obtained for the glycosides.

The unsaturation degree was also calculated from the NMR spectra. Palm oil has higher unsaturation degree compared to palm kernel oil. This is because palm oil contains ~40% of C18:1 alkyl chain. Polyunsaturated compounds were also found in palm oil but not in palm kernel oil. Looking at the glycosides spectra, the unsaturation content in the glycoside based on palm kernel oil resembled the starting alcohol mixture. This indicates no selectivity of the synthesis for saturated or unsaturated alcohols. As mentioned previously, the synthesis of palm oil glycosides catalysed by $SnCl_4$ significantly reduced the unsaturated content while BF_3 retained the amount of the initial palm oil alcohol mixture. Table 3.3 shows the average chain length and the unsaturation degree for glycosides based on palm oil and palm kernel oil.

Table 3.3 Average alkyl chain length and unsaturation content in palm oil and palm kernel oil glycosides based on NMR estimation

Compounds		Average alkyl chain length (carbon)	Unsaturation degree (%)	
			Unsat.	Polyunsat.
Palm oil	Reduced	17	60	10
	Lactoside (BF_3)	17	50	10
	Lactoside ($SnCl_4$)	17	20	~2
	Maltoside (BF_3)	17	60	10
	Maltoside ($SnCl_4$)	17	12	0
Palm kernel oil	Reduced	14	15	0
	Lactoside (BF_3)	14	10	0
	Maltoside (BF_3)	14	15	0

The iodine titration supports the result of the NMR analysis of the unsaturation degree of the compounds. The NMR spectra indicated an approximate 10% loss of unsaturation degree for palm oil and palm kernel oil upon reduction. Table 3.4 shows the iodine value (IV) for each compound. The IV of palm oil and palm kernel oil was comparable with the literature data. From the calculation, there was no loss of unsaturated compound in reduced oil compared to the initial oil for both palm oil and palm kernel oil. The iodine titration results correspond to the calculated value from NMR analysis. Due to the limited amount of palm oil (BF_3) and palm kernel oil derived glycosides, the experiment could not be carried out.

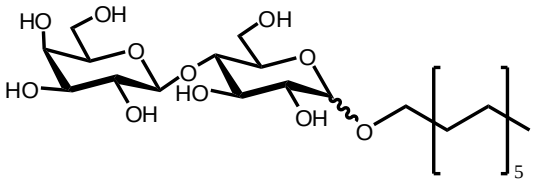
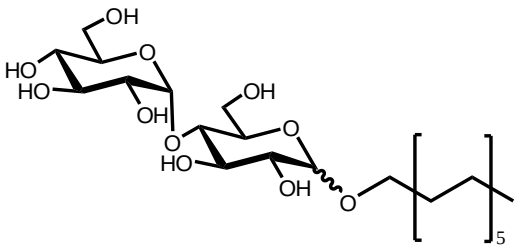
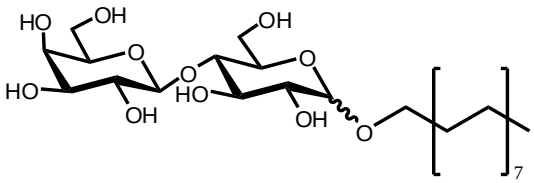
Table 3.4 Experimental and literature iodine value (IV) for palm oil and palm kernel oil

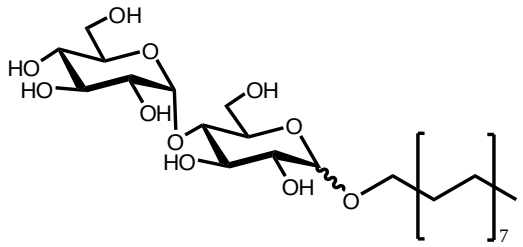
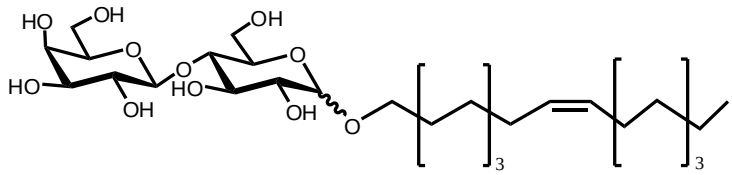
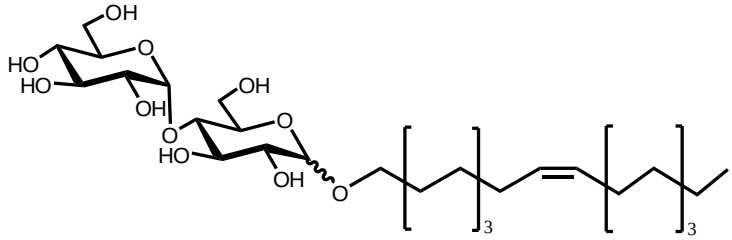
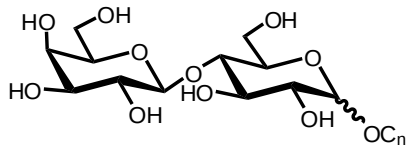
Compounds	IV	Double bond/ mol (based on titration)	NMR based data
Palm Oil	59.0	0.7	0.7
Reduced Palm Oil	65.0	0.6	0.6
Maltoside Palm Oil ($SnCl_4$)	13.0	0.3	0.2
Lactoside Palm Oil ($SnCl_4$)	10.0	0.2	0.2
Palm Kernel Oil	17.0	0.4	0.4
Reduced Palm Kernel Oil	20.0	0.4	0.2
Palm Oil (olein) (Azmil Haizam, et al., 2008)	56.8	-	-
Palm Kernel Oil (Pocklington, 1990)	18.2	-	-

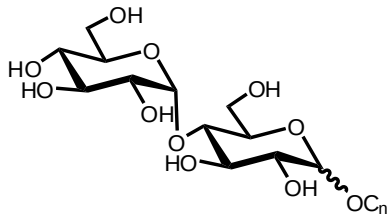
3.1.3 Compound notation

Table 3.5 shows the overview of structures, the chemical names and abbreviations that will be used from here on.

Table 3.5 Compound notations for lactosides and maltosides mixtures

Compounds	Compound Notation	Comment
1-Dodecyl 4-*O*-(β-D-galactopyranosyl)-D-glucopyranoside 	Lacto-C12(α>β)	α-dominant
	Lacto-C12(α≈β)	Balance α/β
	Lacto-C12(α<β)	β-dominant
1-Dodecyl 4-*O*-(α-D-glucopyranosyl)-D-glucopyranoside 	Malto-C12(α>β)	α-dominant
	Malto-C12(α<β)	β-dominant
1-Hexadecyl 4-*O*-(β-D-galactopyranosyl)-D-glucopyranoside 	Lacto-C16	

1-Hexadecyl 4-*O*-(α-D-glucopyranosyl)-D-glucopyranoside 	Malto-C16	
Cis-9-octadecenyl 4-*O*-(β-D-galactopyranosyl)-D-glucopyranoside 	Lacto-C18:1-B	Synthesis using BF_3
	Lacto-C18:1-S	Synthesis using $SnCl_4$
9-octadecenyl-4-*O*-(α-D-glucopyranosyl)-D-glucopyranoside 	Malto-C18:1	
Alkyl lactoside 	Lacto-palm-B	Synthesis using BF_3
	Lacto-palm-S	Synthesis using $SnCl_4$
	Lacto-PKO	

Alkyl maltoside 	Malto-palm-B	Synthesis using BF_3
	Malto-palm-S	Synthesis using $SnCl_4$
	Malto-PKO	

3.2 Physical Properties

Discussion in this chapter will be divided according to the influence of stereochemistry of the sugar, especially the anomeric effect, and the chain effect on the properties of thermotropic and lyotropic liquid crystals. The systems under investigations contain mixtures of different alkyl chains and anomers (α/β) of either lactosides or maltosides.

3.2.1 Sugar stereochemical effect

Investigations were performed on the Malto-C12 and the Lacto-C12 series. Mixtures with different α/β ratio were synthesized and their liquid crystal behaviour, both thermotropic and lyotropic, were characterised. Dodecyl glycosides were selected mainly because the disaccharide and alkyl chain was in balance (HLB balance), thus enable many investigations, especially those, which involves solubility in water, such as the determination of critical micelle concentrations (CMC).

As indicated in chapter 1, straight chain glycosides only exhibit one thermotropic liquid crystalline phase, i.e. the smectic A phase. Conclusively only melting (crystal → smectic phase) and clearing (liquid crystal → isotropic liquid) transition temperatures were observed. These temperatures were recorded by optical polarising microscopy (OPM) and differential scanning calorimetry (DSC).

Table 3.6 Summary of melting and clearing point for dodecyl maltosides and lactosides

Compound	OPM		DSC	
	Melting point (ºC)	Clearing point (ºC)	Melting point (ºC)	Clearing point (ºC)
Malto-C12(α>β)	101	199-202	61 (1st m.p.), 81 (2nd m.p.)	199 (1st c.p.), 211 (2nd c.p.)
Malto-C12(α<β)	80-88	214-229	56 (1st m.p.), 66 (2nd m.p.)	235 (1st c.p.), 233 (2nd c.p.)
Lacto-C12(α>β)	162-166	213-216	165	212
Lacto-C12(α<β)	170-172	244-246	171	239
Lacto-C12(α≈β)	148-158	252-257	152	251

Table 3.6 summarises the melting and clearing transitions temperature for dodecyl maltosides and lactosides. It was noted that β-dominant mixtures have higher clearing point compared to α-dominant mixtures. This trend corresponds to the behaviour of pure compounds (Vill et al., 1989). The higher thermal stability of the liquid crystal phase for the β-anomer has been explained based on its linear structure, i.e. β-anomers have a more linear structure compared to α-anomers (von Minden et al., 2000). The same phenomena also applies for a comparison of lactose and maltose; whereas in lactose the two sugar units are 1,4 β-linked, the corresponding in maltose, is 1,4 α- which is less linear, conclusively the clearing points for lactosides are higher than for maltosides. A similar trend was observed for the melting temperature of lactosides, but for maltosides it was the reverse; i.e. Malto-C12(α>β) seemed to have a higher melting temperature. However Vill et al. (1989) and von Minden et al. (2000) reported significant different melting points for both anomers (refer table 3.7). Besides, von Minden et al. (2000) indicated that there was no certain value for the melting transition since the compounds tend to form non-crystalline solids after lyophilisation. Hence the data are probably inconclusive.

Table 3.7 Comparison of Malto C12 anomeric mixtures with pure Malto C12 anomers

Compounds	Melting point (ºC)	Clearing point (ºC)
Malto-C12(α>β)	101	199-202
Malto-α-C12 (Vill et al., 1989)	*82*	205
Malto-α-C12 (von Minden et al., 2000)	?	205
Malto-C12(α<β)	80-88	214-229
Malto-β-C12 (Vill et al., 2000)	*80*	244
Malto-β-C12 (von Minden et al., 2000)	102	245

On the other hand, the mixture containing an (almost) equal amount of α- and β-anomers, Lacto-C12(α≈β), gave an unexpected high result. For Lacto-C12(α≈β), its melting and clear points contradicts expected trend as it does not show any particular pattern when compared to the other lactosides. It was expected to have both transition temperatures between the ranges of the two mixtures. However, its melting point is lower and the clearing point is higher than others.

According to DSC analysis, all mixtures except Malto-C12(α<β) exhibited degradation. Hence the peaks cannot be reproduced at second heating. As for Malto-C12(α<β), the melting point was recorded only on second heating but not on cooling while the clearing point was detected in both modes. The clearing point decreases at the second heating, which is probably due to impurities originating from partial degradation of the compound upon heating. The increase of the melting point for the second heating may reflect the complexity of maltoside melting point, which is probably due to the

formation of different crystals based on environmental conditions. As indicated before, the melting points for maltosides are probably not suitable for trend analysis. However the increase in clearing point as observed in Malto-C12(α>β) thermogram, was unexplainable.

A region of transition at 30-95^{o}C was found in the Lacto-C12(α>β) thermogram, which origin is unknown. Malto-C12(α>β) showed two sets of transition temperatures, probably indicating different transitions for each anomer.

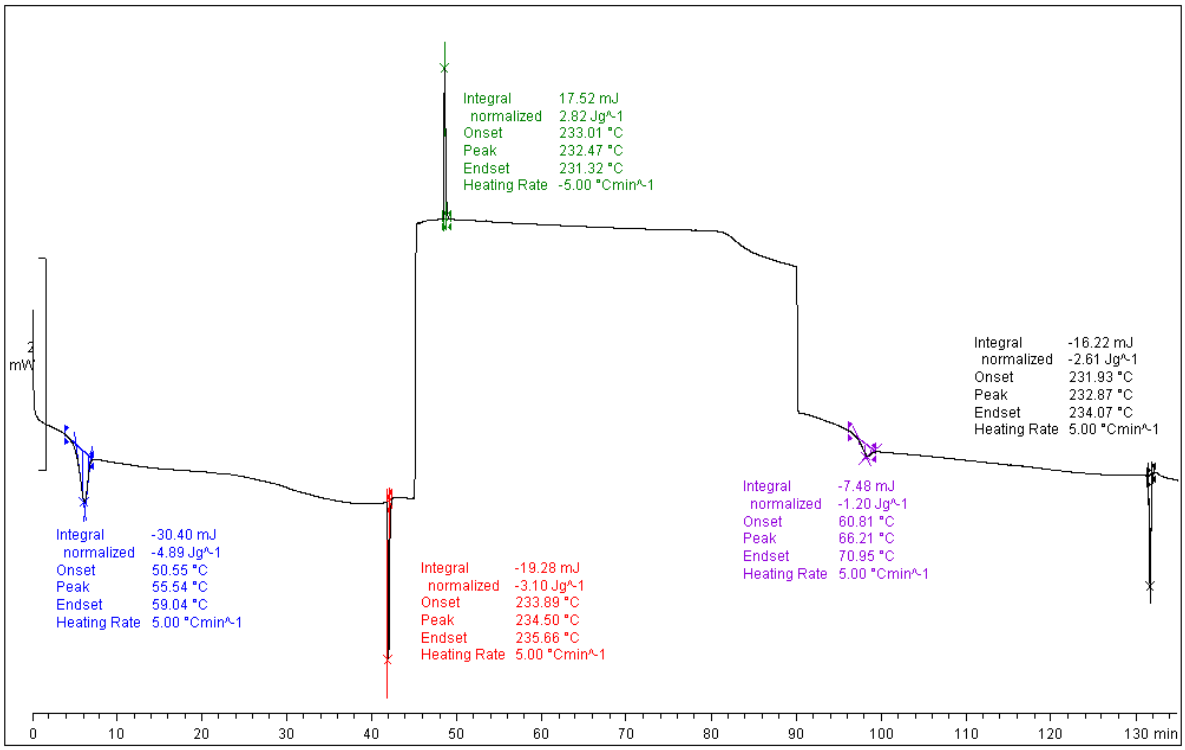


Figure 3.3 DSC thermogram of Malto-C12(α<β)

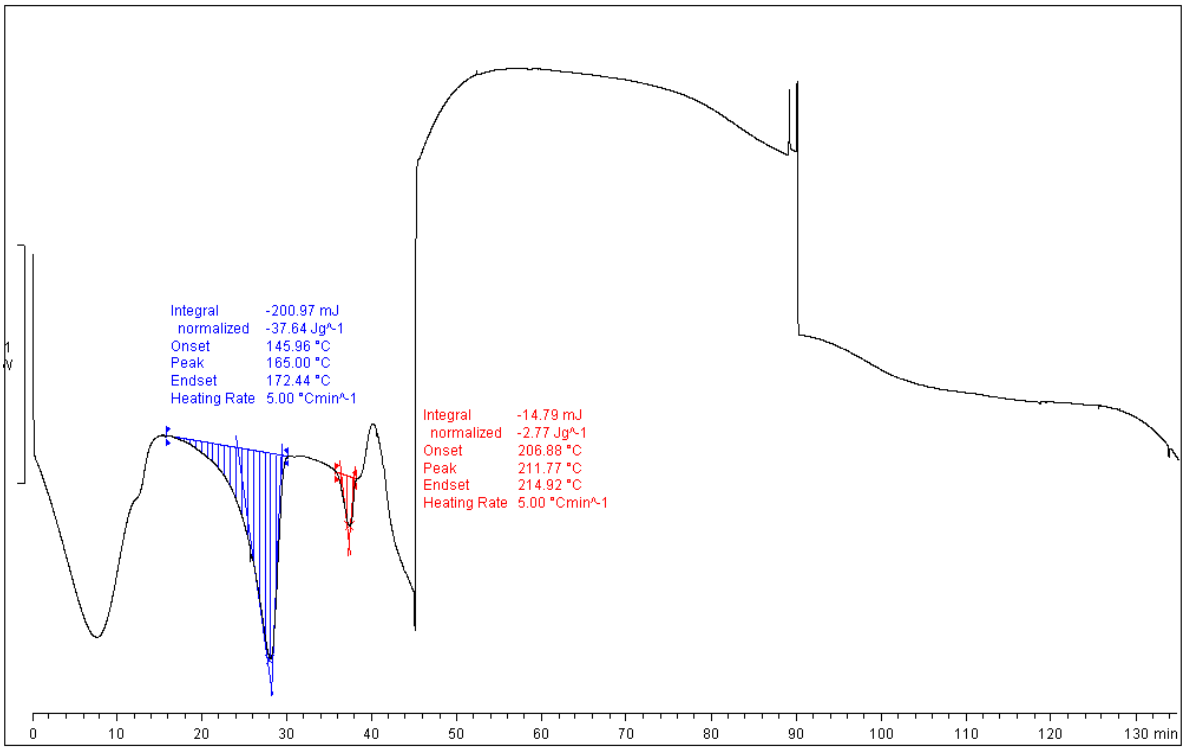


Figure 3.4 DSC thermogram for Lacto-C12(α>β)

Boyd et al. (2000) have investigated Malto-β-C12 using the water penetration scan technique. In the surfactant-water phase diagram Malto-β-C12 displayed in sequence of increasing surfactant concentration; micelles (L_1) → *hexagonal (H_1)* → lamellar (L_α). Both Malto-C12(α>β) and Malto-C12(α<β) mixtures showed similar behaviour. For lactosides, the Lacto-C12(α>β) showed similar phase transitions as maltosides, however only H_I and L_α phases were observed for Lacto-C12(α≈β) and Lacto-C12(α<β). As the β-anomer content increases, the solubility of the compound decreased.

An anisotropic texture with high birefringence (figure 3.5) was observed for every maltoside and lactoside mixture. This phase appeared to be a hexagonal phase, although it did not show the fan-like or mosaic-like texture as observed in Cello-β-C6C2 (Duali Hussen, 2006). However Minamikawa and Hato (2005) made similar observations and

identified the phase as hexagonal (figure 3.6). The phase behaviour of the maltosides and lactosides is shown in table 3.8.

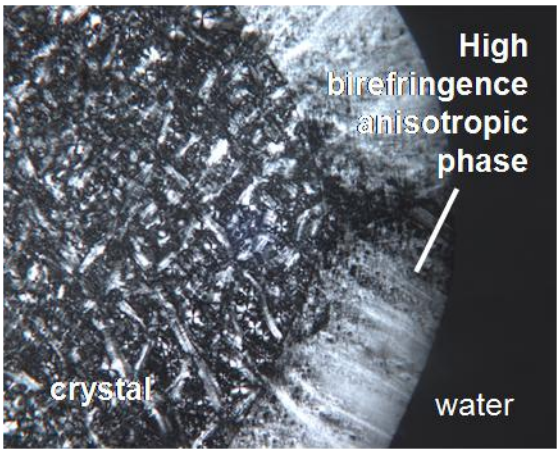


Figure 3.5 Lyotropic phase of Lacto-C12(α>β)

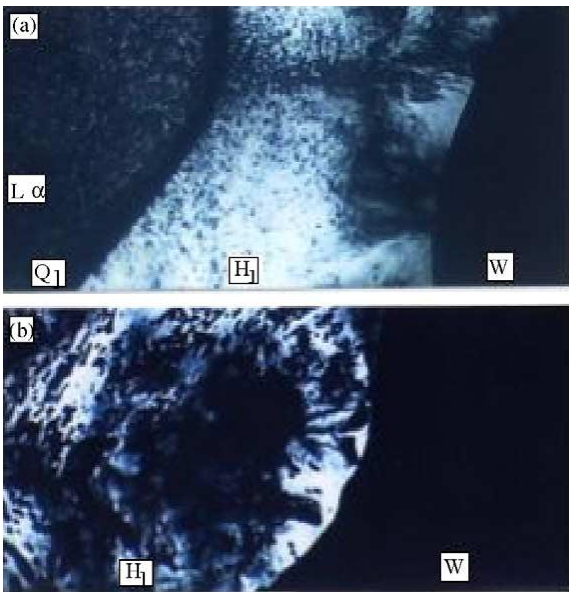


Figure 3.6 Water penetration scans for 1-*O*-geranyl-β-d-maltoside (above) and 1-*O*-geranyl-β-d-maltotrioside (below) (image from Minamikawa and Hato, 2005)

Table 3.8 Phase sequence of dodecyl maltosides and dodecyl lactosides mixtures

Compound	Lyotropic phase
Malto-C12(α>β)	L_1, H_1, L_α
Malto-C12(α<β)	L_1, H_1, L_α
Lacto-C12(α>β)	L_1, H_1, L_α
Lacto-C12(α≈β)	H_1, L_α
Lacto-C12(α<β)	H_1, L_α

In general all mixtures gave similar CMCs (table 3.9). The CMC of maltoside mixtures were comparable with Malto-β-C12 (Boyd et al., 2000 and Niraula et al., 2004). It seems that the anomeric inhomogeneity does not affect the CMC significantly. Having a low CMC shows that these compounds have high degree of hydrophobic effect, thus would perform as good emulsifying agents.

Table 3.9 CMC values of Malto-C12 mixtures and pure Malto-β-C12

Compound	CMC (mM)
Malto-C12(α>β)	0.17
Malto-C12(α<β)	0.15
Malto-β-C12 (Boyd et al., 2000)	0.13
Malto-β-C12 (Niraula et al., 2004)	0.17
Lacto-C12(α>β)	0.15

3.2.2 Chain effect

The chain effect involves two influences, i.e. chain length and chain inhomogeneity. Mixtures of palm oil and palm kernel oil glycosides consist of various alkyl chains in different amounts, thus the mixtures combine chain length effect and chain inhomogeneity effect.

3.2.2.1 Chain length effect

Koeltzow and Urfer (1984) have suggested that liquid crystal transition temperatures (crystal → smectic) are mostly dependent on the alkyl chain length. Addition of more carbons to the alkyl chain increases the intermolecular interactions in both crystal and liquid crystal state (Boyd et al., 2000). The results shown by the mixtures somehow contradicted the statement above. The melting points for lactosides were similar or only slightly increased, whereas the maltosides, due to the formation of non-crystalline solids as stated in 3.2.1 are not discussed. However, the clearing points for both glycosides increase with additional carbons in the chain. For lactosides the increase is small compared to the maltosides. However the data for lactoside are affected by the anomeric effect, therefore no longer only associated with the chain length. The transition temperature trend was more or less in agreement with literature data. Based on results, the clearing point seems to depend on the alkyl chain length whereas the melting point appears to be determined by the sugar and does not show significant chain effects.

The presence of unsaturation in the alkyl chain decreases the clearing temperature of both Malto-C18:1 and Lacto-C18:1. The double bond has been described as a disturbance in the alkyl chain region, lowering the van der Waals interactions by preventing close packing (von Minden et al., 2000). No melting point could be

determined for Malto-C18:1 since the compound only formed a glass. The same behaviour was also observed for Malto-β-C18:1 (von Minden et al., 2000).

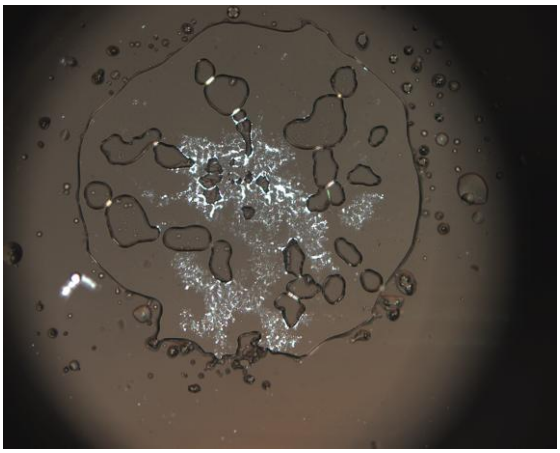
In case of both Lacto-C18:1-B and Lacto-C18:1-S, an isotropic phase was observed along with the smectic phase. A possibility may be the coexistence of a monotropic cubic phase. Von Minden et al. (2000) have reported the occurrence of a monotropic cubic phase for Lacto-α-C18 and Lacto-β-C18. However, due to since the lack of literature data for pure lacto C18:1, the phenomenon was not confirmed. The OPM observation indicates a huge range for the clearing temperature since the mixtures started to clear at around 150°C until it became completely isotropic at 175°C or higher depending on sample size. This probably indicates decomposition of the compounds as the temperature rises, thus no certain value for the clearing point can be given. The clearing temperature is expected to be higher. The DSC thermogram of both compounds supports the degradation assumption since the compounds show a continuous heat effect in the range of 160-175ºC. Table 3.10 shows the thermotropic properties of these glycosides whereas table 3.11 displays literature data of relevant glycosides.

Table 3.10 Thermotropic properties of maltosides and lactosides for various chain lengths

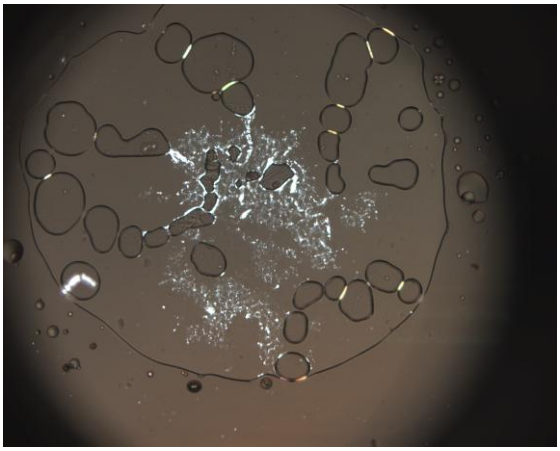
Compound	OPM		DSC	
	Melting point (ºC)	Clearing point (ºC)	Melting point (ºC)	Clearing point (ºC)
Malto-C12(α<β)	80-88	214-229	56	235
Malto-C16	73-76	261-265	61	268
Malto-C18:1	glass	217-221	59	234
Lacto-C12(α≈β)	148-158	252-257	152	251
Lacto-C16	154-166	262	156	252
Lacto-C18:1-B	143-146	152-189	149	~160
Lacto-C18:1-S	143-146	151-192	147	~175

Table 3.11 Literature data of various alkyl chain length for maltosides and lactosides, mainly from von Minden et al. (2000) unless mentioned otherwise

Compound	Melting point (ºC)	Clearing point (ºC)
Malto-β-C12	102	245
Malto-β-C14	107	264
Malto-β-C18	106	274
Malto-β-C18:1	glass	267
Lacto-β-C14 (Vill, 1989)	183	246
Lacto-α-C14 (Vill, 1989)	173	>249
Lacto-β-C18	183	284
Lacto-α-C18	181	260



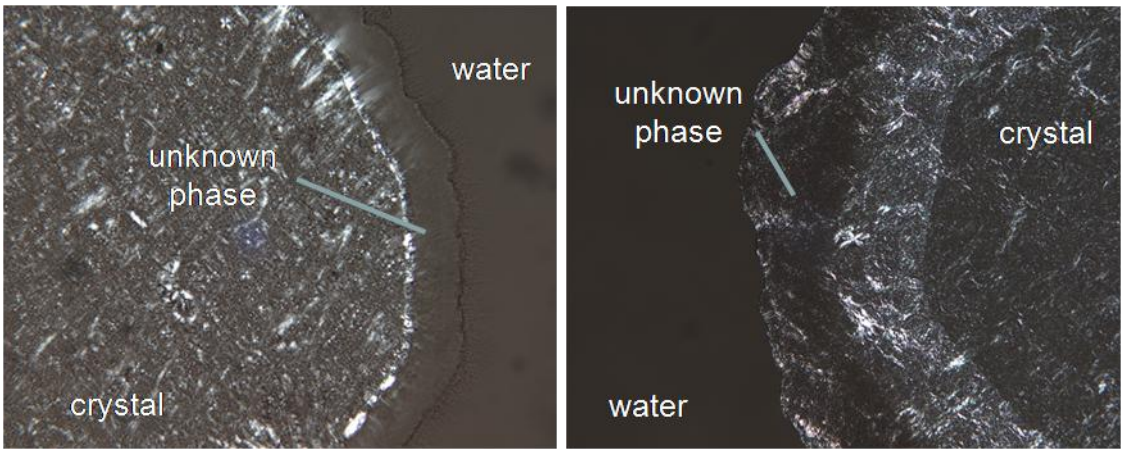
(a)



(b)

Figure 3.7 Lacto-C18:1 at (a) 146ºC and (b) 152ºC

The lyotropic behaviour of dodecyl maltoside and lactoside was already discussed earlier. Increase in alkyl chain length decreases the solubility of the compounds, hence both Malto-C16 and Lacto-C16 did not dissolved. However an interesting phase was observed for both Malto-C16 and Lacto-C16. At higher water concentration region, an isotropic phase was found (figure 3.8). The phase might be assumed as a cubic phase due to the viscosity. Although the phase in Lacto-C16 has completely no texture, Malto-C16 still retained some texture. Possibly these two phases are not of same kind.



(a) (b)

Figure 3.8 Water penetrations of (a) Lacto-C16 and (b) Malto-C16

Malto-C18:1 also exhibited an interesting phase with low anisotropy. The phase appeared at concentrations lower than the hexagonal phase in form of a thin band (figure 3.9). Because it appeared almost isotropic, it might be identified as a micellar cubic phase. However the phase might also be a lyotropic cholesteric phase. Von Minden et al. (2000) observed a possible lyotropic phase for Malto-β-C18:1 which was described as anisotropic phase appearing at higher concentration than the columnar, H_I phase. Based on the criteria, the phase observed has high possibility to be a lyotropic cholestric phase.

Although the unsaturation content of Lacto-C18:1-B and Lacto-C18:1-S were different, they showed similar lyotropic behaviour. Lacto-C18:1-B and Lacto-C18:1-S differs in unsaturation content due to the $SnCl_4$ based reaction of the double bond, as mentioned in 3.1.1.1. However, the solubility increases for Lacto-C18:1-B since it contains more unsaturated compound, and unsaturated compounds increase the water solubility of the mixtures. Figure 3.10 shows the water penetration of Lacto-C18:1-S. The lyotropic behaviour of these mixtures are summarised in table 3.12.

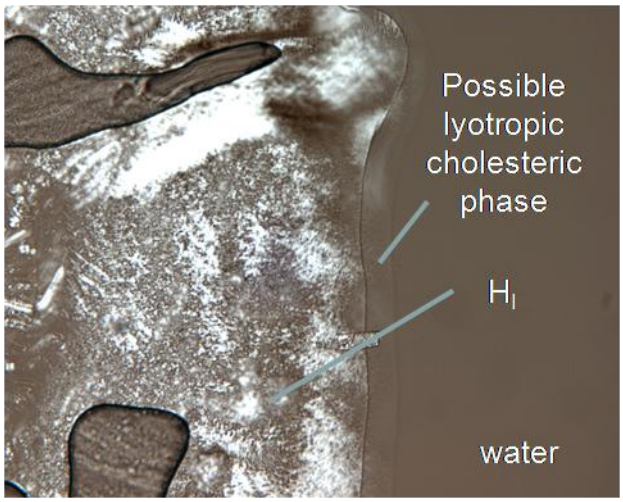


Figure 3.9 Possible lyotropic cholesteric phase in Malto-C18:1

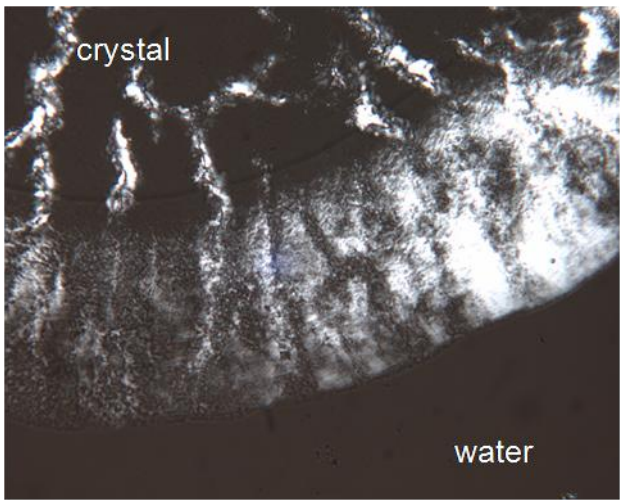


Figure 3.10 Water penetration of Lacto-C8:1-S

Table 3.12 Lyotropic behaviour of maltoside and lactoside mixtures with various alkyl chains

Compound	Lyotropic phase
Malto-C12(α<β)	L_1, H_1, L_α
Malto-C16	unknown, H_1+L_α
Malto-C18:1	L_1, *ch*, H_1, L_α
Lacto-C12(α≈β)	H_1, L_α
Lacto-C16	unknown, H_1+L_α
Lacto-C18:1-B	L_1, H_1, L_α
Lacto-C18:1-S	L_1, H_1, L_α

3.2.2.2 Chain inhomogeneity effect

Palm oil and palm kernel oil are mixtures of various alkyl chains. The fatty acid composition in these oils is different, and that reflects the characteristic of the oil. The fatty acid composition of palm oil and palm kernel oil was already summarised in chapter 1.3.1.

Both Malto-palm-S and Malto-PKO possessed similar thermotropic behaviour reflecting comparable unsaturation contents. The clearing points of both mixtures are low compared to other alkyl chain maltosides. This is due to the presence of unsaturated content in both mixtures. From the chain length study, it was noted that unsaturation decreases the clearing point. The results of Malto-palm-B confirmed the statement. With highest unsaturation content the clearing point is expectedly lowest.

On the contrary, the thermotropic trend in palm oil lactosides was unexpected. Although Lacto-palm-B has a higher unsaturation degree, the clearing point was higher than that of Lacto-palm-S. The result may be explained by the effect of a different α/β ratio,

because Lacto-palm-B is more β-dominant than Lacto-palm-S. The Lacto-PKO showed a clearing point similar to the saturated components. The melting points were more or less the same than those for single chain compounds. As indicated before the chain length does not have a significant effect on the melting point.

The DSC thermogram of Malto-palm indicates degradation of the compound upon heating starting at ~185°C. The Malto-PKO, however, showed a reproducible clearing on cooling and second heating. A slight depression of the temperature for the second heating indicates minor degradation upon clearing, but the degradation is not as significant as for Malto-palm. Malto-PKO also displayed higher clearing point for the second heating as observed in Malto-C12(α>β) which may be from similar cause. The DSC data for both lactosides mixtures match the observations obtained by OPM. Table 3.13 shows the thermotropic properties of maltosides and lactosides based on palm oil and palm kernel oil whereas table 3.14 shows thermotropic properties of maltosides and lactosides with various alkyl chain length.

Table 3.13 OPM and DSC data for thermotropic properties of maltoside and lactoside mixtures based on palm and palm kernel oil

Compound	OPM		DSC	
	Melting point (ºC)	Clearing point (ºC)	Melting point (ºC)	Clearing point (ºC)
Malto-palm-B	108-111	137-144	~60	~180
Malto-palm-S	Glass	199-200	54	~185
Malto-PKO	Glass	195-202	40 (1^{st} heating), 61 (2^{nd} heating)	218 (1^{st} heating), 223 (2^{nd} heating)
Lacto-palm-B	157-159	235-255	157	240
Lacto-palm-S	165	195-196	163	190
Lacto-PKO	162-163	237-241	161	250

Table 3.14 Summary of thermotropic properties for maltosides and lactosides with various alkyl chain lengths

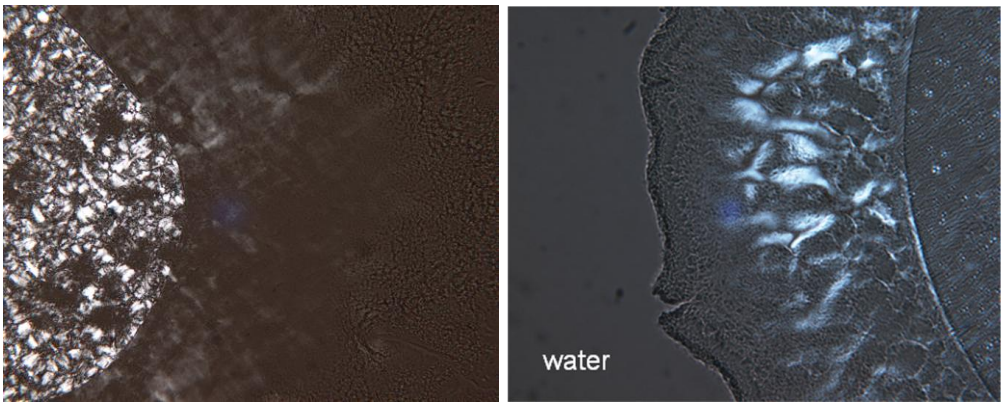
Compound	OPM		DSC	
	Melting point (ºC)	Clearing point (ºC)	Melting point (ºC)	Clearing point (ºC)
Malto-C12(α>β)	101	199-202	61 (1^{st} m.p.), 81 (2^{nd} m.p.)	199 (1^{st} c.p.), 211 (2^{nd} c.p.)
Malto-C12(α<β)	80-88	214-229	56	234.5
Malto-C16	73-76	261-265	61	268
Malto-C18:1	Glass	217-221	59	234
Lacto-C12(α>β)	162-166	213-216	165	212
Lacto-C12(α≈β)	148-158	252-257	152	251
Lacto-C12(α<β)	170-172	244-246	171	239
Lacto-C16	154-166	262	156	252
Lacto-C18:1-B	143-146	152-189	149	~160
Lacto-C18:1-S	143-146	151-192	147	~175

It is indeed a challenge to identify the lyotropic phases of palm oil glycosides. Since the mixture consisted of various alkyl lactosides, involving different chains and anomeric configurations, this multicomponent system showed different phase texture compared to binary system. This lead to some unexplainable texture. As for Lacto-palm-B and Lacto-palm-S, the phases seem to be similar. Both lactosides showed no texture resembling the pattern observed for Lacto-C16 that was accompanying the lamellar phase. In addition, Lacto-palm-S (figure 3.11(b)) somehow displayed a coexistence of a hexagonal and a lamellar ($H_1 + L_\alpha$) phase. This phase was found between the lamellar and the unknown phases. It was not observed for Lacto-palm-B (figure 3.11(a)). The observation was based on the highly birefringent texture observed. The mixtures

behaved more like Lacto-C16 than like Lacto-C18:1. Also the water solubility of these compounds decreases compared to Lacto-C18:1.

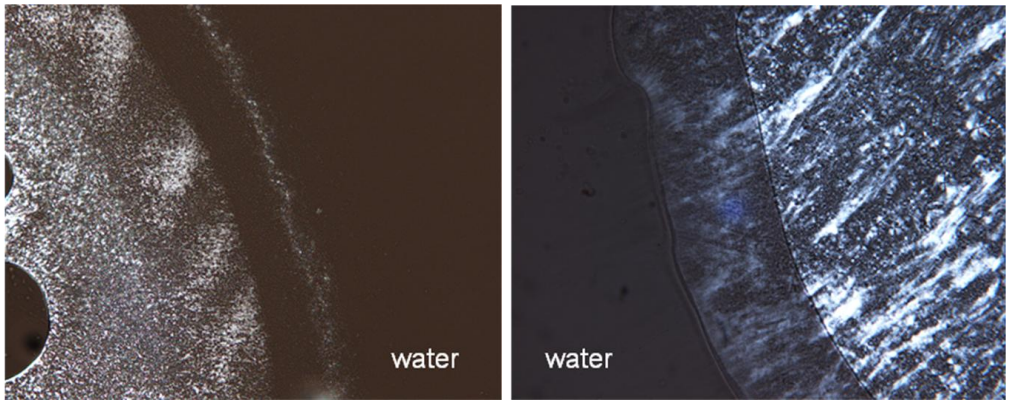
A similar trend was observed for Malto-palm-B and Malto-palm-S (figure 3.12). However the anomeric ratio for these mixtures was not comparable. Thus the anomeric effect adds to a possible chain inhomogeneity effect. The Malto-palm-B showed an additional phase, which could be a cubic phase based on the isotropic appearance and the viscose nature. None of the mixtures showed the possible lyotropic cholestric phase like the one observed in Malto-C18:1.

As for Lacto-PKO and Malto-PKO (figure 3.13), the phase behaviour resembled their palm oil glycosides counterparts. Malto-PKO exhibited a lamellar phase similar to Malto-palm-S, which reflects a similar unsaturation content although the average chain length is different. However Lacto-PKO resembled the behaviour of Lacto-palm-B instead of Lacto-palm-S despite the similarity in the unsaturation content. Table 3.15 summarises the lyotropic behaviour of these mixtures.



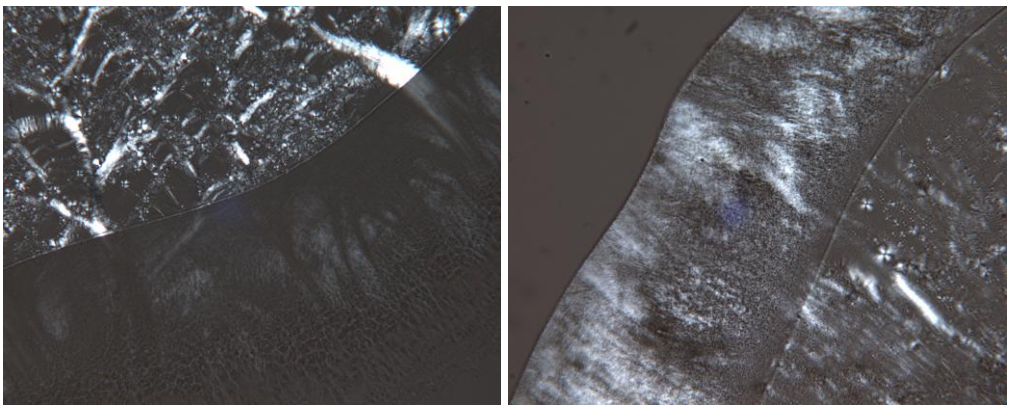
(a) (b)

Figure 3.11 Water penetrations of (a) Lacto-palm-B and (b) Lacto-palm-S



(a) (b)

Figure 3.12 Water penetrations of (a) Malto-palm-B and (b) Malto-palm-S



(a) (b)

Figure 3.13 Water penetrations of (a) Lacto-PKO and (b) Malto-PKO

Table 3.15 Lyotropic behaviour of maltoside and lactoside mixtures with various alkyl chains

Compound	Lyotropic phase
Malto-palm-B	L_1, *Q*, L_α
Malto-palm-S	L_1, L_α
Malto-PKO	L_1, L_α
Lacto-palm-B	Unknown, L_α
Lacto-palm-S	Unknown, H_1, L_α
Lacto-PKO	Unknown, L_α

Several glycolipid mixtures were synthesised from lactose and maltose alcohols derived from palm oil and palm kernel oil. Maltose and lactose were selected as sugar headgroups based on economical aspects. The synthesis involved three steps, which were adapted from Vill et al. (1989). Boron trifluoride (BF_3) and tin tetrachloride ($SnCl_4$) were used to produce different anomeric compositions. BF_3 catalysed reactions produced β-dominant mixtures in short reaction times, whereas longer time led to balanced α/β mixtures. The α-dominant mixtures were synthesized by using $SnCl_4$.

The analysis of the reduced palm oil and palm kernel oil did not show any changes in the alkyl composition. However glycosylation with $SnCl_4$ and unsaturated alcohols, like oleyl alcohol or palm oil based alcohol, led to side reactions which reduced the unsaturation degree significantly. For BF_3 catalysed reactions, the unsaturation content was not affected.

The physicochemical properties of glycolipids are influenced by two major effects, i.e. a stereochemical effect and a chain effect. In order to investigate the thermotropic and lyotropic behaviour, model-compound studies were performed on anomeric mixtures of C_{12}, C_{16} and $C_{18:1}$. These are the major components of palm oil and palm kernel oil. The behaviour of palm oil and palm kernel oil based glycoside can be explained by these effects.

Stereochemistry Effect

β-dominant compounds have higher clearing points than α-dominant compounds. Maltosides tend to form non-crystalline solids, thus melting points could not be observed frequently. As for lactosides, the melting point did not vary significantly.

The anomeric effect did not influence the lyotropic behaviour of the dodecyl maltosides. All mixtures have similar properties to pure Malto-β-C12. However, for

lactosides the solubility in water decreases with dominating β-product. All dodecyl glycosides showed very similar CMCs indicating only neglectable stereochemical effects.

Chain Length Effect

The results show that the clearing point depends on the alkyl chain length, whereas the melting point almost only depends on the headgroup. Unsaturation lowers the thermal stability, thus reducing the clearing point. The water solubility improves with the presence of double bond in the alkyl chain. Malto-C18:1 showed a possible lyotropic cholestric phase, which was not observed for Lacto-C18:1.

Chain Inhomogeneity Effect

The unsaturated compound in the mixture reduces the clearing temperature for palm oil and palm kernel oil maltosides. However this trend is not applicable for lactosides. The behaviour of palm oil based lactoside mixtures is more complicated. Lacto-PKO, on the other hand behaved like saturated lactosides.

Unsaturation influenced the lyotropic behaviour of lactosides and maltosides differently. The lactoside showed no differences upon varying unsaturation content while significant differences were observed for maltosides.

Since most of the lyotropic phases shown by the mixtures are not confirmed, further characterisation should be performed. Detailed investigations on these phases using X-ray (SANS or SWAX) or deuterium NMR could be carried out to better understand the lyotropic behaviour of these mixtures. Besides, studies of the palm oil and palm kernel oil glycosides for bio-related application are recomended.

Attwood, D. and Florence, A.T. (1983). *Surfactant Systems; Their chemistry, pharmacy and biology*, London, New York: Chapman and Hall.

Azmil Haizam, Siew Wai Lin and Ainie Kuntom (2008). Palm-Based Standard Reference Materials for Iodine Value and Slip Melting Point. *Analytical Chemistry Insights* **2008 (3)** 127-133.

Balzer, D. and Lüders, H. (2000). *Nonionic Surfactants, Alkyl Polyglucosides*. New York: Marcel Dekker.

Böcker, T. and Thiem, J. (1989). *Tenside Surfactans Deterg.* **26** 318-324.

Boris B. Niraula, Tham Kok Chun, Hafizrahimi Othman and Misni Misran (2004). Dynamic-interfacial properties of dodecyl-β-D-maltoside and dodecyl-β-D-fructofuranosyl-α-d-glucopyranoside at dodecane/water interface. *Coll. Surf. A Physicochem. Eng. Aspects* **248** 157-166.

Boyd, B.J., Drummond, C.J., Krodkiewska, I. and Grieser, F. (2000). How Chain Length, Headgroup Polymerization, and Anomeric Configuration Govern the Thermotropic and Lyotropic Liquid Crystalline Phase Behaviour and the Air-Water Interfacial Adsorption of Glucose-Based Surfactants. *Langmuir* **16** 7359-7367.

Christ, T., Morgenthaler, W.W. and Pacholec, F. (1994). Laundry Products. In Lange, K.R., *Detergents and Cleaners, A Handbook for Formulators*. Munich; Vienna; New York; Hanser; Cincinnati: Hanser/ Gardner.

Clint, J.H. (1992). *Surfactant Aggregation*, New York: Chapman & Hall.

Corti, M., Cantù, L., Brocca, P. and Del Favero, E. (2007). Self-assembly in glycolipids. *Curr. Opinion Colloid Interface Sci.* **12** 148–154.

D 1959, *Standard Test Method for Iodine Value of Drying Oils and Fatty Acids*, ASTM.

de Vries, R.J. (1984). Utilization of Malaysian Palm Oil and Palm Kernel Oil for Fatty Acids and Derivatives. *J. Am. Oil Chem. Soc.* **61 (2)** 404-407.

Demus, D., Goodby, J., Gray, G.W., Spiess, H.-W. and Vill, V. (1998). *Handbook of Liquid Crystal Vol. 3 High Molecular Weight Liquid Crystal*, Weinheim; New York: Wiley-VCH.

Dörfler, H-D and Göpfert (1999). Lyotropic liquid crystals in binary systems *n-alkyl* glycosides/water. *J. Dispersion Sci Technol.* **23** 35-58.

Dorset, D.L. and Rosenbusch, J.P. (1981). Solid state properties of anomeric 1-O-n-octyl-d-glucopyranosides. *Chem. Phys. Lipids* **29 (4)** 299-307.

Drummond, C.J. and Wells, D. (1998). Nonionic lactose and lactitol based surfactants: comparison of some physic-chemical properties. *Coll. Surf. A Physicochem. Eng. Aspects* **141** 131-142.

Duali Hussen, R.S., 2006. Thesis (MSc.), *Synthesis and Liquid Crystalline Properties of Secondary and Branched Chain of Cellobiosides and Lactosides*, University of Malaya.

Fischer, E. (1893).*Ber. Dtsch. Chem. Ges.* **26** 2400-2412.

Fischer, E. and Helferich, B. (1911). *Justus Liebigs Ann. Chem.* **383** 68-91.

Häntzschel, D., Schulte, J., Enders, S. and Quitzsch, K. (1999). Thermotropic and lyotropic properties of *n*-alkyl-*β*-D-glucopyranoside surfactants. *Phys. Chem. Chem. Phys.* **1** 895-904.

Hashim, R., Abdalla Hashim, H.H., Mohd. Rodzi, N.Z., Duali Hussen, R.S. and Heidelberg, T. (2006). Branched chain glycosides: Enhanced diversity for phase behavior of easily accessible synthetic glycolipids. *Thin Solid Film* **506** 27-35.

Hato, M. (2001). Synthetic glycolipid/water systems. *Curr. Opinion Colloid Interface Sci.* **6** 268-276.

Hato, M., Minamikawa, H., Tamada, K., Baba, T. and Tanabe, Y. (1999). Self-assembly of synthetic glycolipid/water systems. *Adv. Colloid Interface Sci.* **80** 233-270.

Haworth, W.N., Hirst, E.L. and Lyne, R.R., 1937. A Water-Soluble Polysaccharide from Barley Leaves. *Biochem J.* **31 (5)**, 786-788.

Hoffmann, B. and Platz, G. (2001). Phase and aggregation behaviour of alkylglycosides. *Curr. Opinion Colloid Interface Sci.* **6** 171-177.

Holmberg, K. (2001). Natural surfactants. *Curr. Opinion Colloid Interface Sci.* **6** 148-159.

Holmberg, K., Jönsson, B., Kronberg, B. and Lindman, B. (2003). *Surfactants and Polymers in Aqueous Solution 2nd Edition*, England: John Wiley & Sons Ltd.

Jeffrey, G.A. and Wingert, L.M. (1992). Carbohydrate liquid crystals. *Liq. Cryst.* **12** 179-202.

Koeltzow, D.E. and Urfer, A.D. (1984). Preparation and Properties of Pure Alkyl Glucosides, Maltosides and Maltotriosides. *J. Am. Oil Chem. Soc.* **61** 1651-1655.

Lindhorst, T.K. (1999). *Essentials of Carbohydrate Chemistry and Biochemistry*, Weinheim: Wiley-VCH.

Malaysia Palm Oil Board (2007). *Malaysian Oil Palm Statistics 2007*. Malaysia: MPOB.

Malaysian Palm Oil Council (2007). *Fact Sheet; Malaysian Palm Oil*, Malaysia: MPOC and MPOB.

Malaysian Palm Oil Council (2007). *MPOC Annual Report 2007*. Malaysia: MPOC.

Malm, C.J., Tanghe, L.J., Laird, B.C. and Smith G.D. (1953). Relatives rates of acetylation of the hydroxyl groups in cellulose acetate. *J. Am. Chem. Soc*.**75 (1)** 80-84.

Mannock, D.A. and McElhaney, R.N. (2004). Thermotropic and lyotropic phase properties of glycolipid diastereomers: role of headgroup and interfacial interactions in determining phase behavior. *Curr. Opinion Colloid Interface Sci.* **8** 426–447.

Milkereit, G., Garamus, V.M., Veermans, K., Willumeit, R. and Vill, V. (2005). Structures of micelles formed by synthetic alkyl glycosides with unsaturated alkyl chains. *J. Colloid Interface Sci.* **284** 704–713.

Minamikawa, H. and Hato, M. (2005). Headgroup effects on phase behavior and interfacial properties of β-3,7-dimethyloctylglycoside/water systems. *Chem. Phys. Lipids* **134** 151-160.

Mohd. Rodzi, N.Z., 2006. Thesis (MSc.), *Branched Chain Galactosides and Melibiosides: Synthesis and Mesomorphic Properties*, University of Malaya.

Mukerjee, P. and Mysels, K.J. (1971). *Critical Micelle Concentration of Aqueous Surfactant Systems*, Washington, D.C.: Secretary of Commerce.

Nilsson, F., Söderman, O. And Johansson, I. (1998). Four Different C_8G_1 Alkylglucosides, Anomeric Effects and the Influence of Straight vs Branched Hydrocarbon Chains. *J. Colloid Interface Sci.* **203** 131-139.

Nilsson,F., Söderman, O. and Hansson, P. (1998). Physical-Chemical Properties of C_9G_1 and $C_{10}G_1$ *β*-Alkylglucosides. Phase Diagrams and Aggregate Size/ Structure. *Langmuir* **14** 4050-4058.

Nystrom, R. F. and Brown, W. G. (1947). Reduction of organic compounds by lithium aluminum hydride. I. Aldehydes, ketones, esters, acid chlorides and acid anhydrides. *J. Am. Chem. Soc.* **69 (5)** 1197-1199.

Robinson, P.C., Griffin, J.D. and Davidson, M.W. Introduction to Polarized Light Microscopy.
http://www.olympusmicro.com/primer/techniques/polarized/polarizedintro.html

Sakya, P. and Seddon, J.M. (1997). Thermotropic and lyotropic phase behavior of monoalkyl glycosides. *Liq. Cryst.* **23 (3)** 409-424.

Schick, M.J. (1987). Detergency. In Schick, M.J., *Nonionic surfactants*. New York: Marcel Dekker.

Schmitt, T.M. (1992). *Analysis of Surfactants*. New York: Marcel Dekker.

Seddon, J.M., Ces, O., Templer, R.H., Mannock, D.A. and McElhaney, R.N. (2003). Structure and Phase Behaviour of Synthetic Glycolipids. *Mol. Cryst. Liq. Cryst.* **402** 77/[313]-84/[320].

Seddon, J.M., Robins, J., Gulik-Krzywicki, T. and Delacroix, H. (2000). Inverse micellar phases of phospholipids and glycolipids Invited Lecture. *Phys. Chem. Chem. Phys.* **2** 4485-4493.

Shinoda, K., Yamaguchi, T. and Hori, R. (1960). The Surface Tension and the Critical Micelle Concentration in Aqueous Solution of β-D-Alkyl Glucoside and their Mixtures. *J. Phys. Chem.* **34 (2)** 239-241.

Shinoda, K., Yamaguchi, T. and Hori, R. (1989). *Bull. Chem. Soc. Jpn* **34** 237-241.

Shri Singh (2000). Phase transitions in liquid crystals. *Phys. Rep.* **324** 107-269.

Skoog, D.A., Holler, F.J. and Nieman, T.A. (1998). *Principles of Instrumental Analysis 5th Edition*, USA: Brooks/Cole Thomson Learning.

Söderberg, I., Drummond, C.J., Furlong, D. N., Godkin, S. and Matthews, B. (1995). Non-ionic sugar-based surfactants: self assembly and air/water interfacial activity. *Coll. Surf. A Physicochem. Eng. Aspects* **102** 91-97.

Stubenrausch, C. (2001). Sugar surfactants – aggregation, interfacial and adsorption phenomena. *Curr. Opinion Colloid Interface Sci.* **6** 160-170.

Syed Hussan, Rowe, R., Tiddy, G.J.T. (2001). Surfactant liquid crystals and surfactant chemical structure. In Holmberg, K., *Handbook of Applied Surface and Colloid Chemistry*. New York: John Wiley & Sons.

Tschierske, C. (2001). Micro-segregation, molecular shape and molecular topology - partners for the design of liquid crystalline materials with complex mesophase morphologies. *J. Mater. Chem.* **11** 2647-2671.

Vill, V., Böcker, T., Thiem, J. and Fischer, F. (1989). Studies on liquid-crystalline glycosides. *Liq. Cryst.* **6 (3)** 349-356.

von Minden, H.M., 2000. Thesis (PhD), *Synthesis and mesomorphic properties of glycolipids and neoglycolipids*, University of Hamburg.

von Minden, H.M., Brandenburg, K., Seydel, U., Koch, M.H.J., Garamus, V. Willumeit, R. and Vill, V. (2000). Thermotropic and lyotropic properties of long chain alkyl glycopyranosides. Part II. Disaccharide headgroups. *Chem. Phys. Lipids* **106** 157-179.

von Rybinski, W. and Hill, K. (1998). Alkyl Polyglycosides-Properties and Applications of a new Class of Surfactants. *Angew. Chem. Int. Ed.* **37** 1328-1345.

Woolfrey, S.G., Banzon, G.M. and Groves, M.J. (1985). The Effect of Sodium Chloride on the Surface Tension of Sodium Dodecyl Sulfate Solutions. *J. Colloid Interface Sci.* **112 (2)** 583-587.

Zana, R. and Aoudia, M. (1998). Aggregation behavior of sugar surfactants in aqueous solutions: effects of temperature and the addition of non-ionic polymers. *J. Colloid Interface Sci* **206** 158-167.

Zémplen, G. (1929). *Ber. Dtsch. Chem. Ges.* **62** 985.

Zhang, R., Marone, P.A., Thiyagarajan, P., Tiede, D.M. (1999). Structure and molecular fluctuations of *n*-alkyl-*β*-D-glucopyranoside micelles determined by X-ray and neutron scattering. *Langmuir* **15** 7510-7519.

Appendix A

Materials and methods

β-D-lactose (Aldrich) and D(+)-Maltose Monohydrate (Riedel-de Haën) was purchased from Sigma Aldrich. Palm oil alcohol mixtures were obtained from processed palm oil (palm olein) whereas palm kernel oil was obtained from Golden Jomalina Food Industries Sdn. Bhd. Alcohols for model studies were purchased from Fluka and Merck while the catalysts; boron trifluoride (BF_3) and tin tetrachloride ($SnCl_4$) were from Sigma Aldrich. Solvents were AR grade and were used without further purification.

Recycled solvents were used in purification steps. Solvents like dichloromethane, acetonitrile, hexane and n-butanol were redistilled using a rotary evaporator.

General procedures:

i) *General reduction procedure; Synthesis of palm oil and palm kernel oil alcohol mixtures (Nystrom and Brown, 1947).*

A suspension of lithium aluminium hydride ($LiAlH_4$) (33 mmol) in 40 ml diethyl ether was stirred in a two-neck round bottom flask with a closed condenser. A solution of palm oil or palm kernel oil (22 mmol) in 30 ml diethyl ether was added slowly to the suspension through a dropping funnel to avoid vigorous reflux. After the final addition, ethyl acetate, followed by water was added drop wise to destroy excess $LiAlH_4$. An ice-water bath was used to cool the flask due to the heat produced by the reaction. The alcohol mixture was poured into a separating funnel and washed with 100 ml cold aqueous sulphuric acid (10%). After evaporation of diethyl ether, the alcohol mixture was obtained.

ii) *General peracetylation procedure; Synthesis of β-peracetylated sugar (Vogel, 1989; Vill et al., 1989).*

A mixture of sodium acetate (121 mmol) and 100 ml acetic anhydride was stirred and heated to reflux. Sugar (58 mmol) was added in small fraction to the hot suspension. The sugar was dissolved into clear solution because of the exothermic reaction. After all sugar was added, the solution was further heated at 120°C for an hour, and the solution was poured into ice-water and stirred until sticky white solid was formed. It was washed with distilled water for several times until a non-sticky white solid was formed. The product was recrystallized from ethanol. Yield is 75%.

iii) *General glycosidation procedure (Vill, et al., 1989; von Minden, et al. 2000)*

The peracetylated sugar (2.7 mmol) and the alcohol (2.5 mmol) were dissolved in 10 ml dichloromethane and were stirred in a closed apparatus at room temperature. The catalyst, i.e. boron trifluoride or tin tetrachloride (3.5 mmol) was injected into the solution and the reaction was left for the period of time (depending on the parameters applied). The solution was poured into saturated sodium bicarbonate (Na_2CO_3) solution and the organic layer was washed two times with water. For reactions with tin tetrachloride, the solution has to be filtered through wet celite to remove the tin oxide before the extraction step. The dichloromethane was evaporated and an acetonitrile-hexane extraction was performed to remove unreacted alcohol. The acetonitrile layer was collected and evaporated to get the product.

*iv) General deacetylation procedure (*Zémplen, 1929).

Peracetylated glycolipids were dissolved in 50 ml methanol. A catalytic amount of sodium methoxide was added to induce a basic medium. The solution was stirred for 3 hours and the progress of the reaction was monitored by TLC. After the reaction completed, methanol was evaporated and the product was separated by an extraction with n-butanol and water. A small amount of diluted sulphuric acid was added to neutralize the excess sodium methoxide. The organic layer was collected and the solvent was evaporated to obtain the product. The product was dried in a vacuum oven at 50°C for 48 hours.

v) Iodine titration

A mixture of 10 ml Wijs solution, ~0.2 g sample and 8 ml carbon tetrachloride (CCl_4) was placed in a conical flask with stopper and stored for one hour in the dark. Then 8 ml of aqueous potassium iodide solution (0.9 M) followed by 40 ml water was added to the mixture. The liberated iodine was titrated with sodium thiosulphate (0.02M) until the yellowish colour almost disappears. Afterwards a few drops of starch solution were added as an indicator and the titration continues until the blue colour disappears. A blank was also prepared by adding the same amount of Wijs solution and carbon tetrachloride except no sample was added and was titrated until the end point. The IV was calculated by the following equation:

$$IV\,(g/100\,g) = \frac{12.69C(V_1 - V_2)}{m}$$

Where,

C = concentration of sodium thiosulphate solution (mol dm^{-3})

V_1 = volume (ml) of sodium thiosulphate solution used for the blank titration

V_2 = volume (ml) of sodium thiosulphate solution used for the sample titration

m = mass (g) of sample

Appendix B

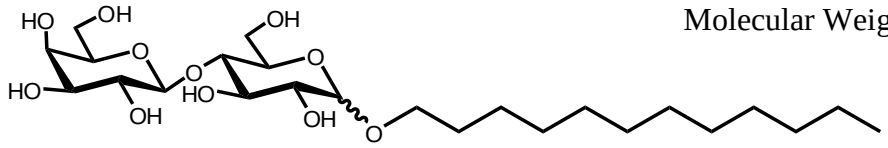
Compound preparations and chemical analsyis

1-Dodecyl 4-*O*-(β-D-galactopyranosyl)-D-glucopyranoside

Lacto-C12

Chemical Formula: $C_{24}H_{46}O_{11}$

Molecular Weight: 510.62 $gmol^{-1}$



α-dominant (α/ β ratio: 7:1)

9.5 g (14.9 mmol) β-lactose octaacetate and 3.1 g (16.8 mmol) 1-dodecanol were reacted with 3.3 ml (18.2 mmol) tin tetrachloride using the general glycosylation procedure. Reaction period is 7 hours. Yield is 9.4 g.

The crude Lacto-C12-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 3.9 g (51%).

^{1}H-NMR (400 MHz, C_5D_5N): δ = 5.26 (d, 88%H, αH-1, J = 3.6 Hz), 5.12 (d, 1H, H-1'), 5.02 (d, 12%H, βH-1, J = 7.9 Hz), 3.50-4.81 (m, bulk sugar signals), 1.50-1.79 (m, 2H, $-CH_2-$), 1.24-1.35 (m, 18H, $-CH_2-$), 0.87 (t, 3H, CH_3)

β-dominant (α/ β ratio: ~1:20)

9.5 g (14.9 mmol) β-lactose octaacetate and 3.1 g (16.8 mmol) 1-dodecanol were reacted with 2.3 ml (18.2 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 7 hours. Yield is 7.9 g.

The crude Lacto-C12-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 3.0 g (40%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 4.79 (d, 5%H, αH-1, J = 4.0 Hz), 4.37 (d, 1H, H-1'), 4.28 (d, 95%H, βH-1, J = 8.0 Hz), 3.24-3.88 (m, bulk sugar signals), 1.60-1.64 (m, 2H, -CH_2-), 1.29-1.37 (m, 18H, -CH_2-), 0.89 (t, 3H, CH_3)

balance (α/ β ratio: 1:1)

Compound obtained from Dr. Thorsten Heidelberg. Yield 54%.

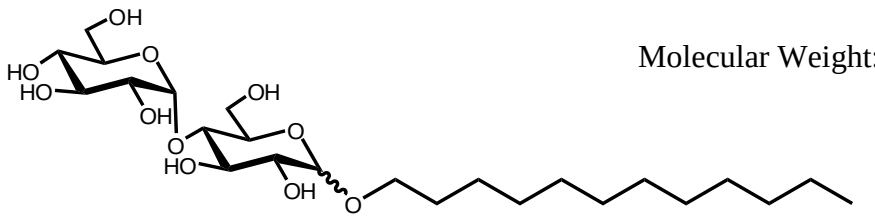
^{1}H-NMR (400 MHz, CD_3OD): δ = 4.68 (d, 50%H, αH-1, J = 3.8 Hz), 4.37 (d, 1H, H-1'), 4.28 (d, 50%H, βH-1, J = 7.8 Hz), 3.22-3.76 (m, bulk sugar signals), 1.45-1.55 (m, 2H, -CH_2-), 1.21-1.40 (m, 18H, -CH_2-), 0.81 (t, 3H, CH_3)

1-Dodecyl 4-*O*-(α-D-glucopyranosyl)-D-glucopyranoside

Malto-C12

Chemical Formula: $C_{24}H_{46}O_{11}$

Molecular Weight: 510.62 $gmol^{-1}$



α-dominant (α/ β ratio: 7:1)

9.5 g (14.9 mmol) β-maltose octaacetate and 3.1 g (16.8 mmol) 1-dodecanol were reacted with 3.3 ml (18.2 mmol) tin tetrachloride using the general glycosylation procedure. Reaction period is 6 hours. Yield is 9.4 g.

The crude Malto-C12-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 4.5 g (60%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.15 (d, 1H, H-1'), 4.77 (d, 88%H, αH-1, J = 3.7 Hz), 4.27 (d, 12%H, βH-1, J = 7.8 Hz), 3.25-3.90 (m, bulk sugar signals), 1.62-1.64 (m, 2H, -CH_2-), 1.30-1.37 (m, 18H, -CH_2-), 0.90 (t, 3H, CH_3)

β-dominant (α/ β ratio: 1:5)

15 g (23.6 mmol) β-maltose octaacetate and 5.1g (27.0 mmol) 1-dodecanol were reacted with 3.7 ml (29.0 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 6 hours. Yield is 14.2 g.

The crude Malto-C12-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 8.3 g (69%).

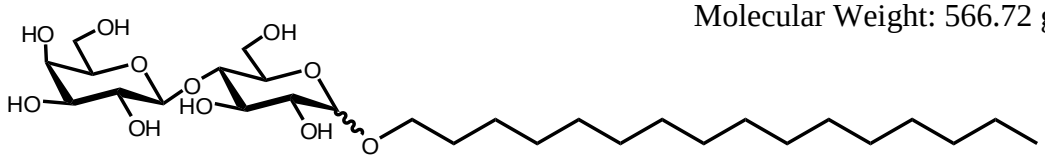
^{1}H-NMR (400 MHz, $CDCl_3$): δ = 5.17 (d, 1H, H-1'), 4.77 (d, 17% H, αH-1, J = 3.8 Hz), 4.27 (d, 83%H, βH-1, J = 7.8 Hz), 3.20-3.92 (m, bulk sugar signals), 1.51-1.65 (m, 2H, -CH_2-), 1.29-1.37 (m, 18H, -CH_2-), 0.90 (t, 3H, CH_3)

1-Hexadecyl 4-*O*-(β-D-galactopyranosyl)-D-glucopyranoside

Lacto-C16 (α/ β ratio: 2:1)

Chemical Formula: $C_{28}H_{54}O_{11}$

Molecular Weight: 566.72 $gmol^{-1}$



1.7g (2.7 mmol) β-lactose octaacetate and 0.6g (2.5 mmol) 1-hexadecanol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 48 hours. Yield is 1.6 g.

The crude Lacto-C16-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.3 g (17%).

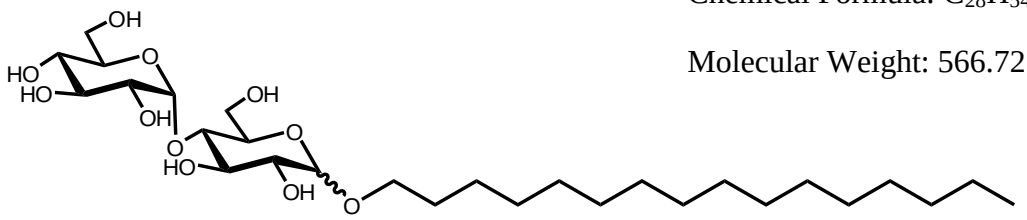
^{1}H-NMR (400 MHz, CD_3OD): δ = 4.77 (d, 67%H, αH-1, J = 3.8 Hz), 4.36 (d, 1H, H-1'), 4.28 (d, 33%H, βH-1, J = 7.9 Hz), 3.30-3.87 (m, 14H, bulk sugar signals), 1.62-1.64 (m, 2H, H-β), 1.29-1.37 (m, 28H, -CH_2-), 0.89 (t, 3H, CH_3)

1-Hexadecyl 4-*O*-(α-D-glucopyranosyl)-D-glucopyranoside

Malto-C16 (α/ β ratio: 1:6)

Chemical Formula: $C_{28}H_{54}O_{11}$

Molecular Weight: 566.72 $gmol^{-1}$



1.7g (2.7 mmol) β-maltose octaacetate and 0.6g (2.5 mmol) 1-hexadecanol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 6 hours. Yield is 1.9 g.

The crude Malto-C16-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.5 g (32%).

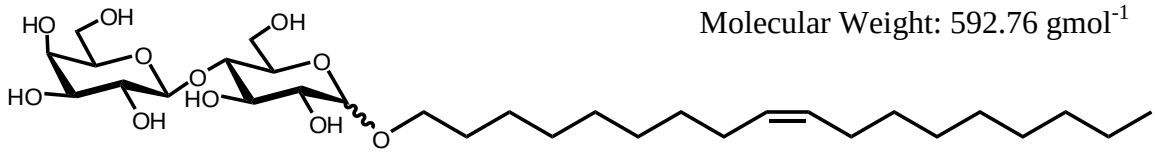
^{1}H-NMR (400 MHz, CD_3OD): δ = 5.15 (d, 1H, H-1'), 4.76 (d, 14%H, αH-1, J = 4.0 Hz), 4.26 (d, 86%H, βH-1, J = 8.0 Hz), 3.19-3.91 (m, bulk sugar signals), 1.58-1.63 (m, 2H, -CH_2-), 1.28-1.37 (m, 28H, -CH_2-), 0.89 (t, 3H, CH_3)

Cis-9-octadecenyl 4-*O*-(β-D-galactopyranosyl)-D-glucopyranoside

Lacto-C18:1

Chemical Formula: $C_{30}H_{56}O_{11}$

Molecular Weight: 592.76 $gmol^{-1}$



Lacto-C18:1-B (α/ β ratio: 1:1)

1.7g (2.7 mmol) β-lactose octaacetate and 0.6g (2.5 mmol) cis-9-octadecen-1-ol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 48 hours. Yield is 1.9 g.

The crude Lacto-C18:1-B-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.6 g (36%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.33 (m, 2H, -CH=), 4.76 (d, 50%H, αH-1, J = 3.8 Hz), 4.36 (d, 1H, H-1'), 4.27 (d, 50%H, βH-1, J = 7.8 Hz), 3.24-3.83 (m, bulk sugar signals), 2.03 (m, 4H, -CH_2-CH=), 1.61 (m, 2H, -CH_2-), 1.29-1.31 (m, 22H, -CH_2-), 0.88 (t, 3H, CH_3)

Lacto-C18:1-S (α/ β ratio: 2:1)

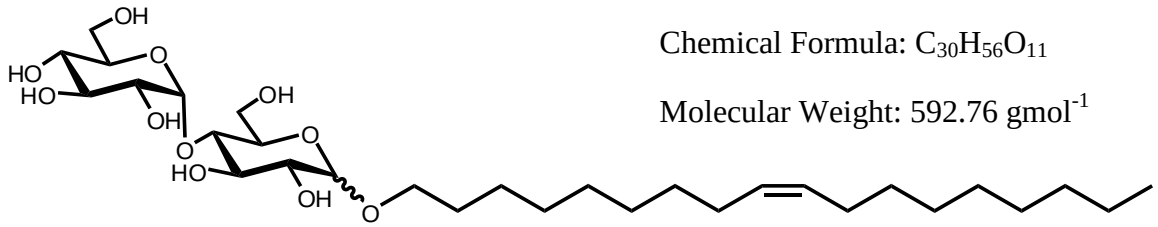
1.7g (2.7 mmol) β-lactose octaacetate and 0.6g (2.5 mmol) cis-9-octadecen-1-ol were reacted with 0.3 ml (2.5 mmol) tin tetrachloride using the general glycosylation procedure. Reaction period is 7 hours. Yield is 1.5 g.

The crude Lacto-C18:1-S-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.2 g (16%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.35 (m, 0.8H, -CH=), 4.77 (d, 67%H, αH-1, J = 3.7 Hz), 4.37 (d, 1H, H-1'), 4.28 (d, 33%H, βH-1, J = 7.9 Hz), 3.24-3.89 (m, bulk sugar signals), 1.97-2.04 (m, 1.6H, -CH_2-CH=), 1.60-1.64 (m, 2H, -CH_2-), 1.29-1.32 (m, 22H, -CH_2-), 0.90 (t, 3H, CH_3)

Cis-9-octadecenyl 4-*O*-(α-D-glucopyranosyl)-D-glucopyranoside

Malto-C18:1 (α/ β ratio: 1:6)



Chemical Formula: $C_{30}H_{56}O_{11}$

Molecular Weight: 592.76 $gmol^{-1}$

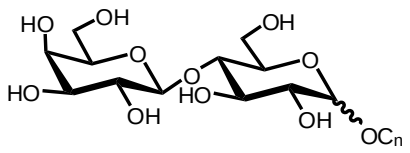
1.7g (2.7 mmol) β-maltose octaacetate and 0.6g (2.5 mmol) cis-9-octadecen-1-ol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 6 hours. Yield is 1.7 g.

The crude Malto-C18:1-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.3 g (22%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.33-5.39 (m, 2H, -CH=), 5.16 (d, 1H, H-1'), 4.77 (d, 14%H, αH-1, J = 3.7 Hz), 4.27 (d, 86%H, βH-1, J = 7.8 Hz), 3.20-3.90 (m, bulk sugar signals), 1.97-2.05 (m, 4H, $-CH_2-CH=$), 1.52-1.66 (m, 2H, $-CH_2-$), 1.29-1.32 (m, 22H, $-CH_2-$), 0.90 (t, 3H, CH_3)

Alkyl lactoside based palm oil

Lacto-palm



Lacto-palm-B (α/ β ratio: 1:1)

1.7 g (2.7 mmol) β-lactose octaacetate and 0.6 g (2.5 mmol) palm oil alcohol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etheaate using the general glycosylation procedure. Reaction is 24 hours. Yield is 1.8 g.

The crude Lacto-palm-B-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.4 g (23%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.33-5.36 (m, 1H, -CH=), 4.77 (d, 50%H, αH-1, J = 4.0 Hz), 4.37 (d, 1H, H-1'), 4.28 (d, 50%H, βH-1, J = 7.6 Hz), 3.25-3.89 (m, bulk sugar signals), 2.76-2.79 (m, 0.1H, =CH-CH_2-CH=) 2.02-2.05 (m, 2H, -CH_2-), 1.61-65 (m, 2H, -CH_2-), 1.29-1.33 (m, 23H, -CH_2-), 0.90 (t, 3H, CH_3);

Lacto-palm-S (α/ β ratio: 2:1)

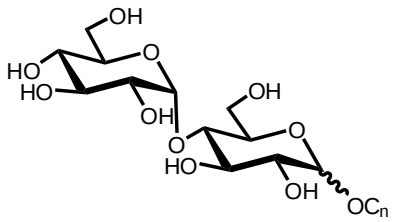
9.5 g (14.9 mmol) β-lactose octaacetate and 4.1 g (16.8 mmol) palm oil alcohol were reacted with 3.3 ml (18.2 mmol) tin tetrachloride using the general glycosylation procedure. Reaction period is 7 hours. Yield is 2.8 g.

The crude Lacto-palm-S-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 1.3 g (25%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.35-5.39 (m, 0.3H, -CH=), 4.77 (d, 67%H, αH-1, J = 3.6 Hz), 4.36 (d, 1H, H-1'), 4.29 (d, 33%H, βH-1, J = 7.6 Hz), 3.20-3.85 (m, bulk sugar signals), 1.98-2.03 (m, 0.7H, -CH_2-), 1.48 -1.76 (m, 2H, -CH_2-), 1.29 (m, 23H, -CH_2-), 0.89 (t, 3H, CH_3)

Alkyl maltoside based palm oil

Malto-palm



Malto-palm-B (α/ β ratio: 1:6)

1.7 g (2.7 mmol) β-maltose octaacetate and 0.6 g (2.5 mmol) palm oil alcohol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 6 hours. Yield is 1.5 g.

The crude Malto-palm-B-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.5 g (31%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.33-5.36 (m, 1.2H, -CH=), 5.15-5.16 (d, 1H, H-1'), 4.77 (d, 14%H, αH-1, J = 3.8 Hz), 4.26 (d, 86%H, βH-1, J = 7.6 Hz), 3.20-3.92 ((m, bulk sugar signals), 2.76-2.79 (m, 0.1H, =CH-CH_2-CH=), 2.01-2.07 (m, 2.5H, -CH_2-CH=), 1.53 -1.64 (m, 2H, -CH_2-), 1.29-1.32 (m, 23H, -CH_2-), 0.90 (t, 3H, CH_3)

Malto-palm-S (α/ β ratio: 2:1)

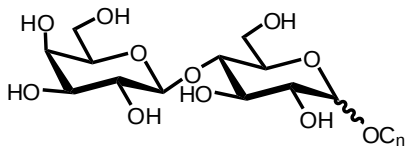
9.5 g (14.9 mmol) β-maltose octaacetate and 4.1 g (16.8 mmol) palm oil alcohol were reacted with 3.3 ml (18.2 mmol) tin tetrachloride using the general glycosylation procedure. Reaction period is 6 hours. Yield is 2.5 g.

The crude Malto-palm-S-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 1.2 g (28%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.33-5.39 (m, 0.2H, -CH=), 5.15-5.17 (m, 1H, H-1'), 4.78 (d, 67%H, αH-1, J = 3.6 Hz), 4.27 (d, 33%H, βH-1, J = 8.0 Hz), 3.21-3.9 (m, bulk sugar signals), 1.98-2.03 (m, 0.5H, -CH_2-), 1.53 -1.64 (m, 2H, -CH_2-), 1.29-1.32 (m, 25H, -CH_2-), 0.90 (t, 3H, CH_3)

Alkyl lactoside based palm kernel oil

Lacto-PKO (α/ β ratio: 1:1)



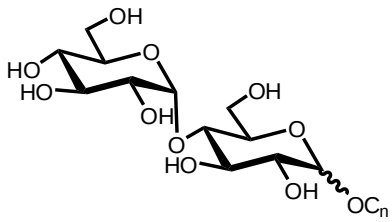
1.7g (2.7 mmol) β-lactose octaacetate and 0.6g (2.5 mmol) palm kernel oil alcohol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 24 hours. Yield is 1.6 g.

The crude Lacto-PKO-OAc peracetylated was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.5 g (35%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.35 (m, 0.2H, -CH=), 4.77 (d, 50%H, αH-1, J = 3.6 Hz), 4.35-4.38 (m, 1H, H-1'), 4.28 (d, 50%H, βH-1, J = 8.0 Hz), 3.23-3.92 (m, bulk sugar signals), 2.02-2.04 (m, 0.5H, -CH_2-), 1.60 -1.65 (m, 2H, -CH_2-), 1.29-1.32 (m, 20H, -CH_2-), 0.90 (t, 3H, CH_3)

Alkyl maltoside based palm kernel oil

Malto-PKO (α/ β ratio: 1:2)



1.7g (2.7 mmol) maltose octaacetate and 0.6g (2.5 mmol) palm kernel oil alcohol were reacted with 0.3 ml (3.5 mmol) boron trifluoride diethyl etherate using the general glycosylation procedure. Reaction period is 6 hours. Yield is 1.8 g.

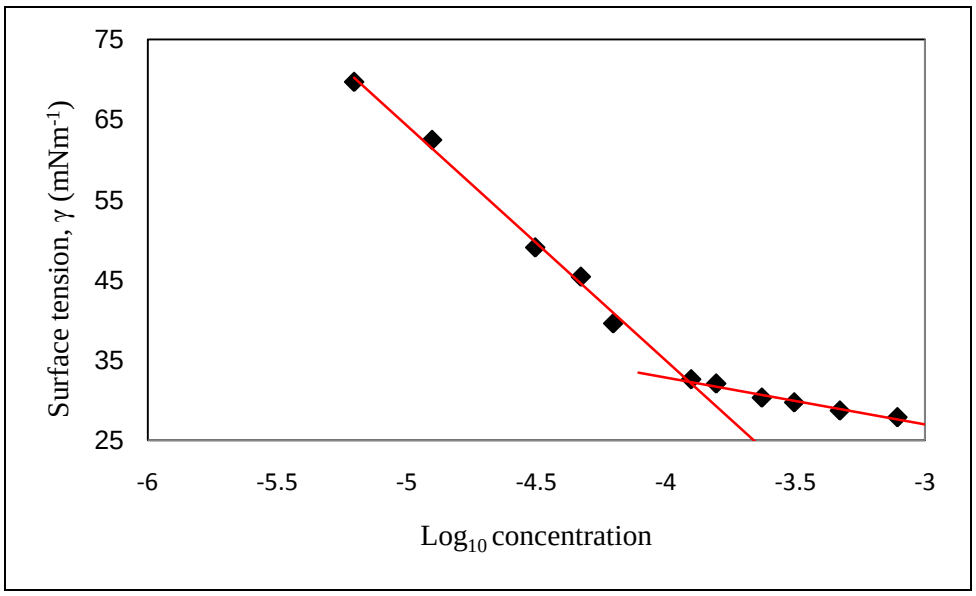
The crude Malto-PKO-OAc was deacetylated according to the general deacetylation method. The reaction takes 3 hours to complete. Yield 0.4 g (33%).

^{1}H-NMR (400 MHz, CD_3OD): δ = 5.35 (m, 0.3H, -CH=), 5.15 (m, 1H, H-1'), 4.77 (d, 33%H, αH-1, J = 3.6 Hz), 4.27 (d, 67%H, βH-1, J = 8.0 Hz), 3.20-3.90 (m, bulk sugar signals), 2.02-2.07 (m, 0.6H, -CH_2-),1.53-1.64 (m, 2H, -CH_2-), 1.29 (m, 20H, -CH_2-), 0.90 (t, 3H, CH_3).

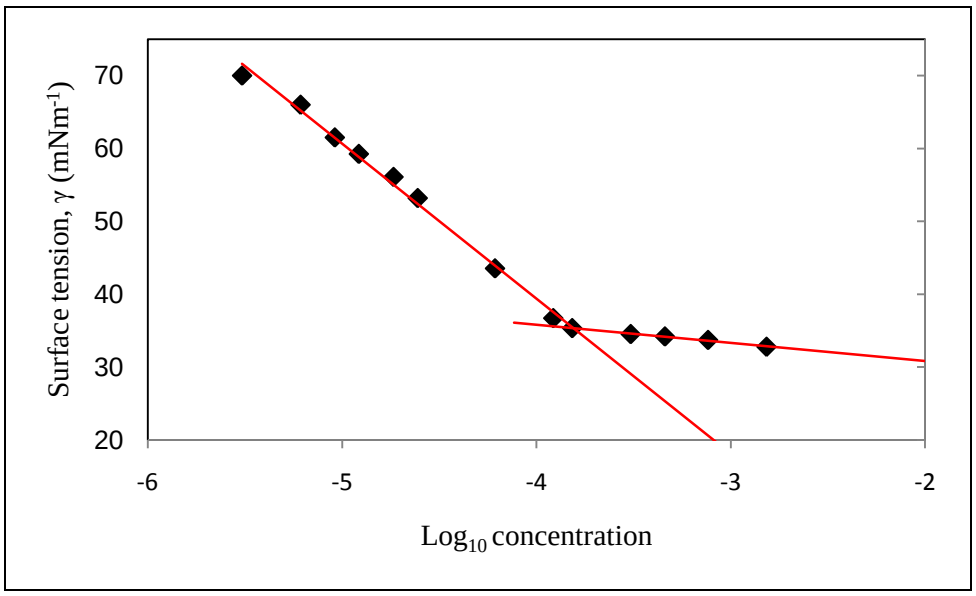
Appendix C

CMC results

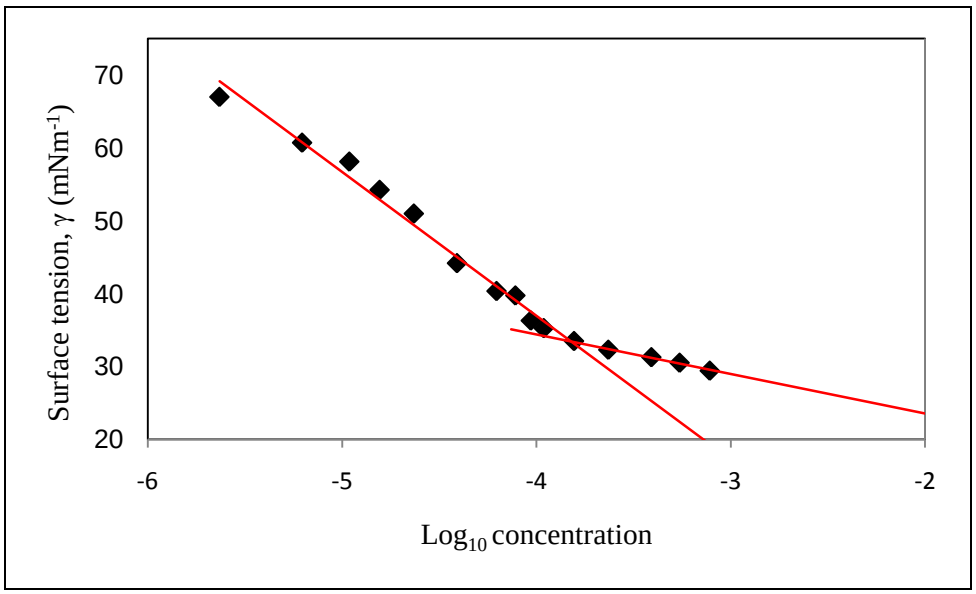
1. Lacto-C12(α>β) CMC: 0.15



2. Malto-C12(α>β) CMC: 0.17



3. Malto-C12(α<β) CMC: 0.15



Appendix D

Risk and safety information

The risk (R) and safety (S) informations of all chemicals used throughout the research.

Name	R	S
1-dodecanol	38, 50	61
1-hexadecanol	36/37/38	22, 24/25
Acetic anhydride	10, 20/22, 34	26, 36/37/39, 45
Acetone	11, 36, 66, 67	9, 16, 26
Acetonitrile	11, 23/24/25	16, 27, 45
Amberlite IR resin	No data available	No data available
Boron trifluoride diethyl etherate	14/15, 34	26, 28, 36/37/39, 45
Carbon tetrachloride	23/24/25, 40, 48/23, 52/53, 59	23, 36/37, 45, 59,61
Chloroform	22, 38, 40, 48/20/22	36/37
Cis-9-octadecen-1-ol	No data available	No data available
Dichloromethane	40	23, 24/25, 36/37
Ethanol	11	7, 16
Diethyl ether	12, 19, 22, 66, 67	9, 16, 29, 33
Ethyl acetate	11	16, 23, 29, 33
Hexane	11, 38, 48/20, 51/53, 62, 65, 67	9, 16, 29, 33, 36/37, 61, 62
Hydrochloric acid	34, 37	26, 36/37/39, 45
Lactose	No data available	No data available
Lithium aluminium hydride	15	7/8, 24/25, 43
Magnesium sulfate	-	-
Maltose	No data available	No data available
Methanol	11, 23/25	7, 16, 24, 45
Palm kernel oil	No data available	No data available
Palm oil	No data available	No data available
Potassium iodide	No data available	No data available
Sodium acetate, anhydrous	-	22, 24/25
Sodium methoxide	11, 14, 34	8, 16, 26, 46, 11, 45
Sodium thiosulfate	No data available	No data available
Tin tetrachloride	34, 37	7/8, 26, 45
Wijs solution	10, 35	23, 26, 36/37/39, 45

Appendix E

List of scientific presentations

Poster Presentations

1. Effects of Chain Inhomogeneity on Physcial Properties of Alkyl Glycosides, 2nd Mathematics and Physical Sciences Graduate Convention (MPSGC), December 2006, National University of Singapore (NUS), Singapore.

2. Model-Studies on Palm Oil based Glycosides, 2nd Penang International Conference for Young Chemists (Penang ICYC) 2008, 18th – 20th June 2008, University of Science Malaysia.

Oral Presentations

3. Stereochemical Effects on Physical Properties of Glycosides Surfactant, 3rd Mathematics and Physical Sciences Graduate Convention (MPSGC), 12th – 14th December 2007, Faculty of Science, University of Malaya.